



Europäisches Patentamt  
European Patent Office  
Office européen des brevets



(11) Publication number:

0 549 040 A1

(12)

## EUROPEAN PATENT APPLICATION

(21) Application number: 92203905.2

(51) Int. Cl.5: C07H 17/08, A61K 31/70

(22) Date of filing: 12.12.92

(30) Priority: 20.12.91 US 811412

(71) Applicant: MERCK & CO. INC.  
126, East Lincoln Avenue P.O. Box 2000  
Rahway New Jersey 07065-0900(US)

(43) Date of publication of application:  
30.06.93 Bulletin 93/26

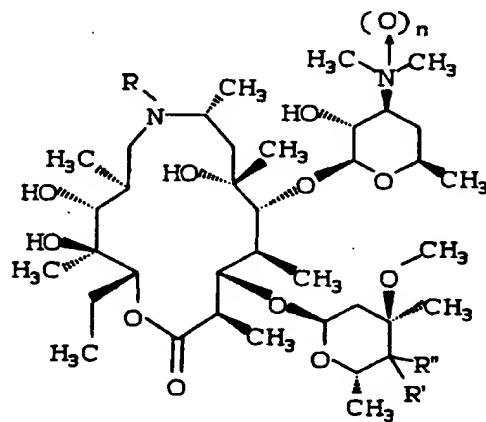
(72) Inventor: Shankaran, Kothandaraman  
391 College Drive  
Edison, NJ 08817(US)  
Inventor: Wilkening, Robert R.  
11 Salter Place  
Maplewood, NJ 07042(US)

(84) Designated Contracting States:  
CH DE FR GB IT LI NL

(74) Representative: Thompson, John Dr. et al  
Merck & Co., Inc. European Patent  
Department Terlings Park Eastwick Road  
Harlow, Essex CM20 2QR (GB)

(54) Methods of making 4" derivatives of 9-deoxy-8a-aza-8a-alkyl-8a-homoerythromycin A.

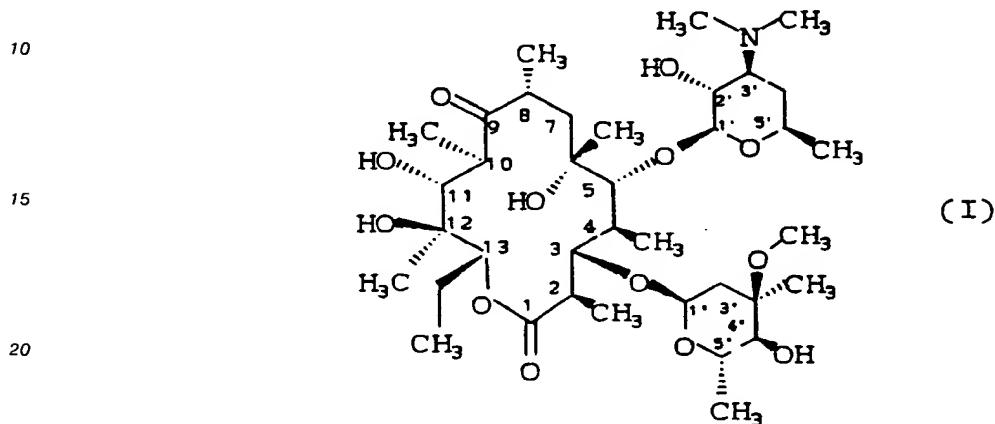
(57) A method of making compounds of the formula:



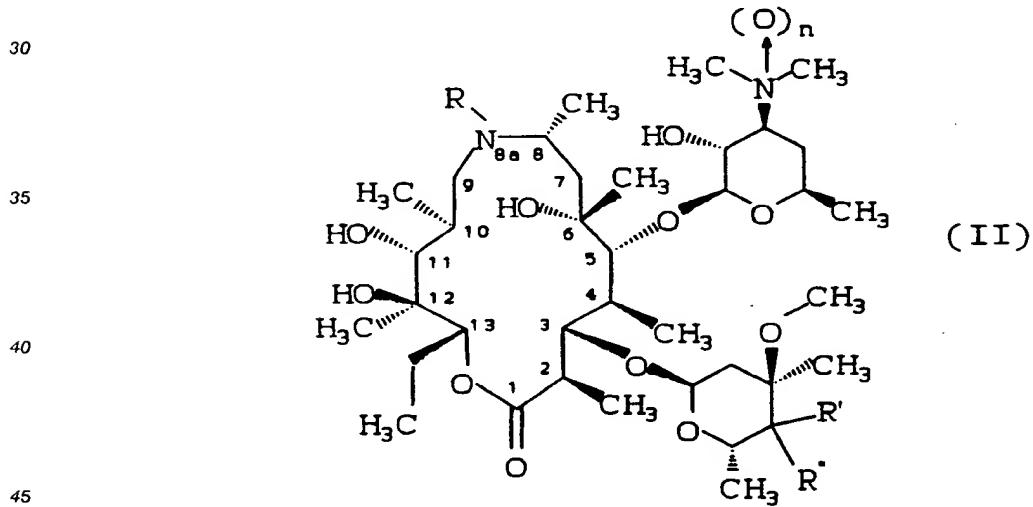
EP 0 549 040 A1

BACKGROUND OF THE INVENTION

The present invention relates to a novel method of making a novel group of chemical compounds having antibacterial activity, which are useful in the therapy of bacterial infections in mammals. More 5 specifically, the invention relates to methods of making derivatives of the well-known macrolide antibiotic, erythromycin A, the compound of the structure:



25 Even more specifically, the invention relates to novel methods of making the compounds of the structure:

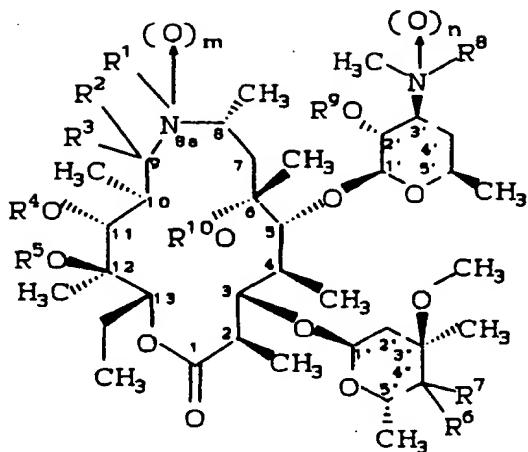


50 wherein R is hydrogen, hydroxyl, C<sub>1-10</sub> alkyl or C<sub>1-10</sub> alkoxy carbonyl C<sub>1-10</sub> alkyl and n is 0 or 1, R' and R" together are oxo or hydroxyimino, and R' and R" independently are hydrogen, hydroxyl or amino substituted by any of hydrogen, alkyl carbonyl, aryl carbonyl, aralkyl carbonyl, alkoxy carbonyl or aralkoxy carbonyl.

The present invention also provides for novel pharmaceutical compositions and methods of their use as antibiotic agents.

55 SUMMARY OF THE INVENTION

The present invention relates to novel methods of making compounds that are macrolides of the following structure:



(III)

and the pharmaceutically acceptable salts and esters thereof, and the pharmaceutically acceptable metal complexes thereof, wherein

20 R<sup>1</sup> is hydrogen,

hydroxy,

formyl,

C<sub>1-10</sub> alkylcarbonyl,

C<sub>1-10</sub> alkoxy

25 C<sub>1-10</sub> alkoxy carbonyl,

aryloxycarbonyl,

C<sub>1-10</sub> aralkoxycarbonyl,

arylsulfonyl,

30 C<sub>1-10</sub> alkylsulfonyl, or

unsubstituted or substituted C<sub>1-10</sub> alkyl wherein said substituent is: 1-3 fluoro, aryl, amino, substituted amino wherein said substituent is formyl or C<sub>1-10</sub> alkylcarbonyl; cyano, isonitrolo, oxo, carboxy, carbamoyl, Br, Cl, mono- or disubstituted amino wherein said substituents are C<sub>1-C10</sub> alkyl or mono- or di-hydroxy-substituted aryl C<sub>1-10</sub> alkyl; nitro, heterocyclyl, heteroaryl, substituted carbamoyl wherein the nitrogen atom is mono or di-substituted with C<sub>1-10</sub> alkyl; aryloxy, arythio, C<sub>1-10</sub> alkoxy carbonyl, hydroxy, C<sub>1-10</sub> alkoxy, mercapto, C<sub>1-10</sub> alkylthio, C<sub>1-10</sub> alkylsulfinyl, C<sub>1-10</sub> alkylsulfonyl, unsubstituted or substituted iminomethyl wherein said substituent is C<sub>1-C10</sub> alkyl; sulfamoyl, unsubstituted or substituted C<sub>1-10</sub> alkylamino wherein said substituent is amino, oxo or formyl and C<sub>1-10</sub> alkylcarbonyl;

40 R<sup>1</sup> and R<sup>10</sup> together are C<sub>1-C3</sub> alkylene optionally substituted by an oxo group;

R<sup>1</sup> and R<sup>4</sup> together are C<sub>1-C3</sub> alkylene optionally substituted by an oxo group

R<sup>2</sup> and R<sup>3</sup> are hydrogen,

C<sub>1-10</sub> alkyl,

aryl

45 R<sup>2</sup> and R<sup>3</sup> together are

oxo and

thiono;

R<sup>4</sup> and R<sup>5</sup> are independently

50 hydrogen and

alkylcarbonyl;

R<sup>4</sup> and R<sup>5</sup> are together

carbonyl

R<sup>6</sup> and R<sup>7</sup> are independently

55 hydrogen,

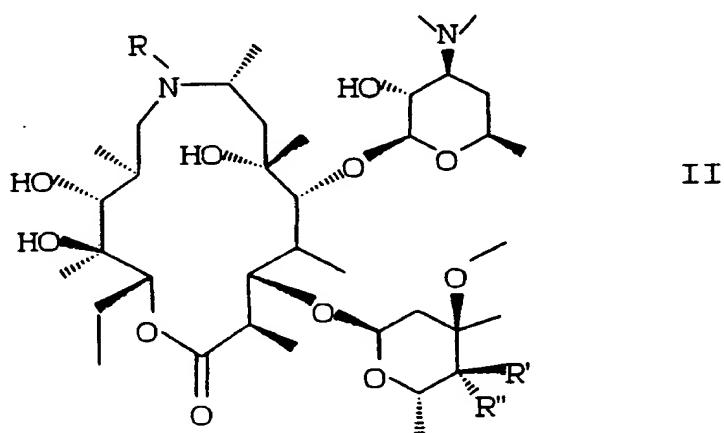
hydroxy,

C<sub>1-10</sub> alkylcarbonyloxy and

-NHR<sup>12</sup> wherein R<sup>12</sup> is hydrogen, C<sub>1-10</sub> alkylcarbonyl, arylsulfonyl; halogen-substituted arylsulfonyl, thiensulfonyl, furansulfonyl, or



5 where  
 X is a connecting bond, O or NH,  
 A is a connecting bond or  $C_1-C_3$  alkylene,  
 10  $R^{13}$  is hydrogen,  $C_1-C_{10}$  alkyl, aryl, aralkyl, heteroaryl, heterocyclyl, or  $C_3-C_7$  cycloalkyl, any of  
 which  $R^{13}$  groups other than hydrogen can be substituted by one or more of halogen, hydroxyl,  
 $C_1-C_3$  alkoxy, cyano, isonitrilo, nitro, amino, mono- or di- $(C_1-C_3)$  alkylamino,  $C_1-C_{10}$  alkylamide,  
 mercapto,  $C_1-C_3$  alkylthio,  $C_1-C_3$  alkylsulfinyl,  
 15  $C_1-C_3$  alkylsulfonyl, arylthio, arylsulfinyl, sulfamoyl, arylsulfonyl, carboxy, carbamoyl,  $C_1-C_3$   
 alkylcarbonyl,  $C_1-C_3$  alkoxy carbonyl or  $C_1-C_3$ -substituted-halogen.  
 20  $R^6$  and  $R^7$  are together  
 oxo and  
 hydroxyimino;  
 $R^8$  is  
 methyl, aralkoxy carbonyl, and  
 25  $R^9$  is  
 hydrogen,  
 $C_1-C_{10}$  alkylcarbonyl, and  $C_1-C_{10}$   
 alkoxy carbonyl and aralkoxy carbonyl;  
 30 m and n are independently integers of zero or one;  
 and said metal complex is taken from the group consisting of  
 copper,  
 zinc,  
 cobalt,  
 35 nickel and  
 cadmium.  
 A most preferred group of compounds made by the method of the present invention is that of the  
 following formula



50 wherein R is hydrogen, hydroxyl,  $C_1-C_{10}$  substituted or unsubstituted alkyl, or  $C_1-C_{10}$  alkoxy carbonyl  $C_1-C_{10}$  alkyl,  $R'$  and  $R''$  together are oxo or hydroxyimino, and  $R'$  and  $R''$  independently are hydrogen, hydroxyl or amino substituted by any of hydrogen,  $C_1-C_{10}$  alkylcarbonyl,  $C_1-C_{10}$  alkoxy carbonyl, aryl carbonyl, aryl  $C_1-C_{10}$  alkylcarbonyl or aryl  $C_1-C_{10}$  alkoxy carbonyl and n is 0 or 1, and the pharmaceutically acceptable salts and esters thereof.

55 The most preferred compounds made by the method of the present invention are 4'' aminated derivatives of 9-deoxy-8a-aza-8a-alkyl-8a-homoerythromycin A, that is, when  $R'$  or  $R''$  are amino or substituted amino. The method of obtaining such derivatives comprises the steps of

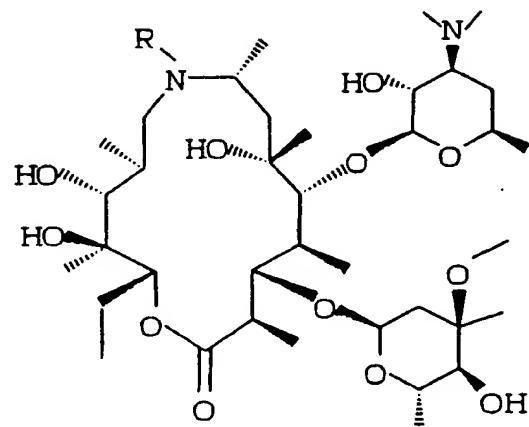
(a) oxidizing a compound of the formula

5

10

15

20

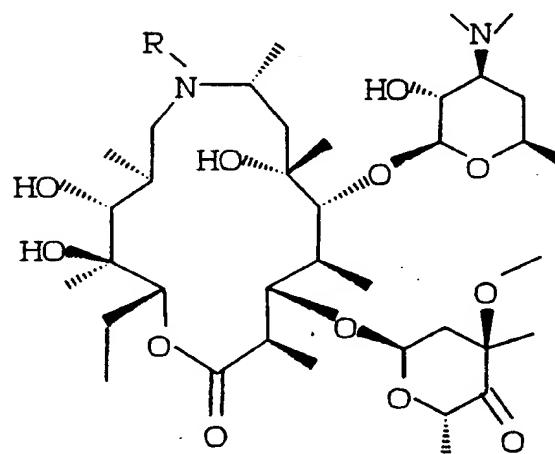


25

30

35

40

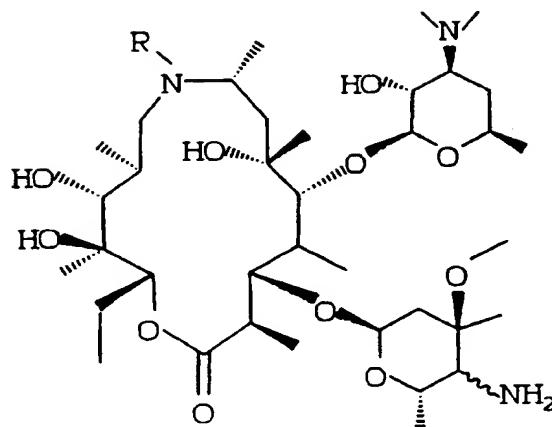


45

50

55

5



10

15

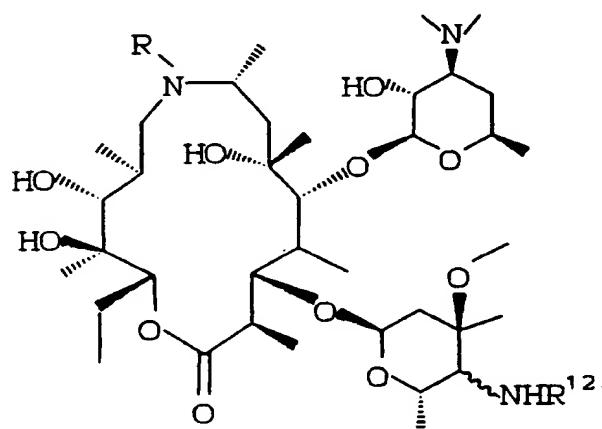
Additionally, the product of step (b) can be substituted at the amino moiety of the cladinose sugar to produce a product of the formula

20

25

30

35



40 where  $R^{12}$  is  $C_{1-10}$  alkyl,  $C_{1-10}$  alkyl carbonyl,  $C_{1-10}$  alkoxy carbonyl, aryl carbonyl, aryl  $C_{1-10}$  alkyl carbonyl or aryl  $C_{1-10}$  alkoxy carbonyl.

Such salts are generally prepared as acid addition salts by combining the compound of formula II with one to three equivalents of an appropriate acid in an inert solvent. The salt is then recovered by solvent evaporation or by filtration if the salt precipitates spontaneously, or by precipitation using a co-solvent or a 45 non-polar co-solvent followed by filtration.

Representative salts and esters include the following:

50

55

5	Acetate	Lactobionate
	Benzenesulfonate	Laurate
	Benzoate	Malate
	Bicarbonate	Maleate
	Bisulfate	Mandelate
10	Bitartrate	Mesylate
	Borate	Methylsulfate
	Bromide	Mucate
	Calcium Eddate	Napsylate
	Camsylate	Nitrate
	Carbonate	Oleate
	Chloride	Oxalate
	Clavulanate	Pamoate (Embonate)
15	Citrate	Palmitate
	Dihydrochloride	Pantothenate
	Eddate	Phosphate/diphosphate
	Edisylate	Polygalacturonate
	Estolate	Salicylate
20	Esylate	Stearate
	Ethylsuccinate	Subacetate
	Fumarate	Succinate
	Glceptate	Tannate
	Gluconate	Tartrate
	Glutamate	Teoclare
25	Glycolylarsanilate	Tosylate
	Hexylresorcinate	Triethiodode
	Hydrabamine	Valerate
	Hydrobromide	
	Hydrochloride	
30	Iodide	
	Iothionate	
	Lactate	

35 DETAILED DESCRIPTION OF THE INVENTION

The term "pharmacologically effective amount" shall mean that amount of a drug or pharmaceutical agent that will elicit the biological or medical response of a tissue, system or animal that is being sought by 40 a researcher or clinician.

The term "antibiotically effective amount" shall mean that amount of an antibiotic compound that will achieve a level of antibacterial activity at the site of infection that is sufficient to inhibit the bacteria in a manner that allows the host organism to overcome the infection.

The term "alkyl" shall mean cyclic or linear straight or branched chain alkane, alkene or alkyne of one 45 to ten carbon atoms unless some specific number is given (e.g., C<sub>1</sub>-C<sub>3</sub>) with one or more degrees of unsaturation.

The term aryl shall include phenyl and fluorenyl.

The term heteroaryl shall mean unsaturated cyclic structures having one or four heteroatoms which can be O, N or S.

50 The term heterocyclyl shall mean saturated cyclic structures, having one to two heteroatoms which can be O, N or S.

Whenever the terms "alkyl" or "aryl" or either of their prefix roots appear in a name of a substituent (e.g., aralkoxy aryloxyalkyl) they shall be interpreted as including those limitations given above for "alkyl" and "aryl". Designated numbers of carbon atoms (e.g., C<sub>1</sub>-C<sub>10</sub> or C<sub>1</sub>-10) shall refer independently to the 55 number of carbon atoms in an alkyl or cyclic alkyl moiety or to the alkyl portion of a larger substituent in which alkyl appears as its prefix root.

The term "substituted" shall be deemed to include multiple degrees of substitution by a named substituent.

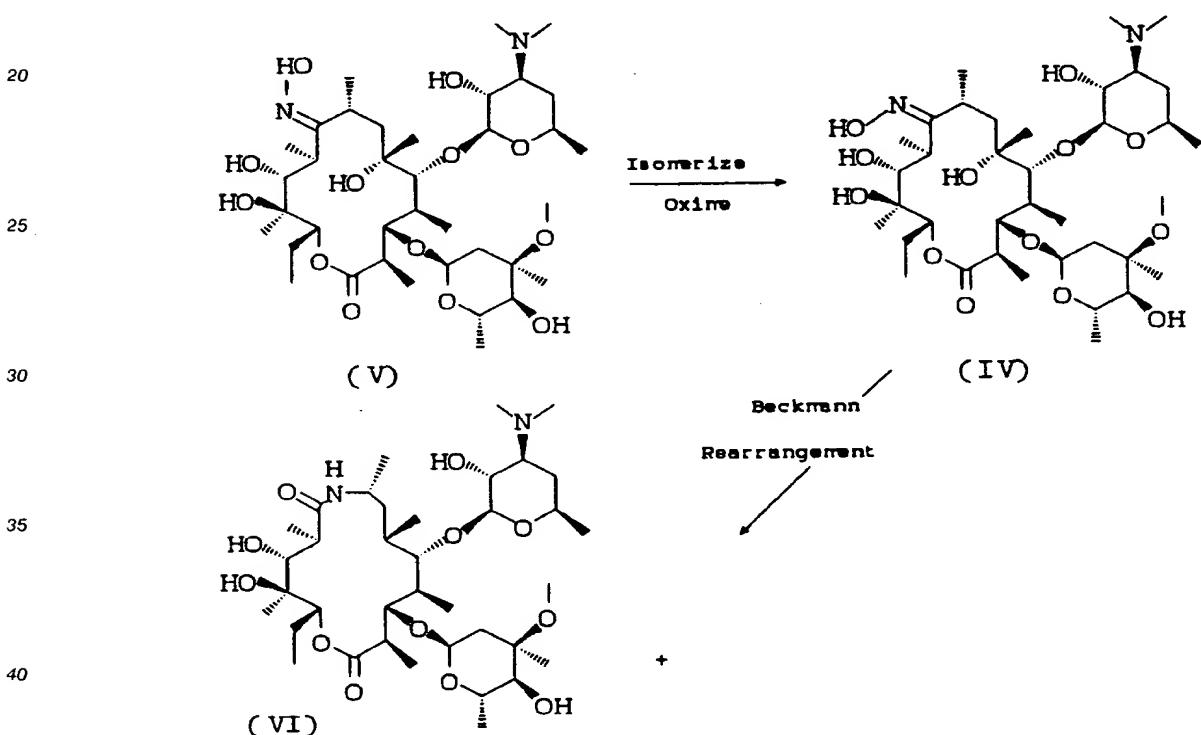
Where multiple substituent moieties are disclosed or claimed, the substituted compound can be independently substituted by one or more of the disclosed or claimed substituent moieties, singly or plurally.

The compounds of formula II can be prepared readily according to the following detailed descriptions and examples or modifications thereof using readily available starting materials, reagents and conventional synthesis procedures. The overall process is illustrated in the following flow charts. In these reactions, it is also possible to make use of variants which are themselves known to those of ordinary skill in this art, but which are not mentioned in greater detail.

10

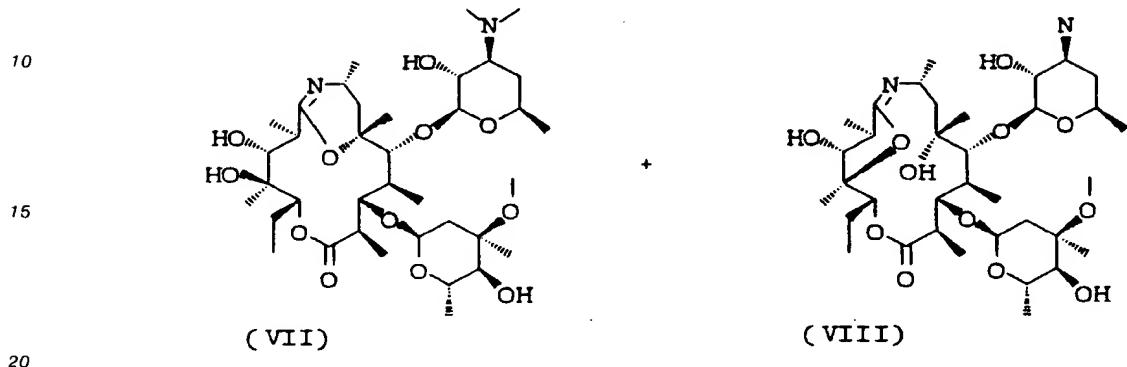
FLOW CHART I

15



FLOW CHART (Cont'd)

5



25

30

35

40

45

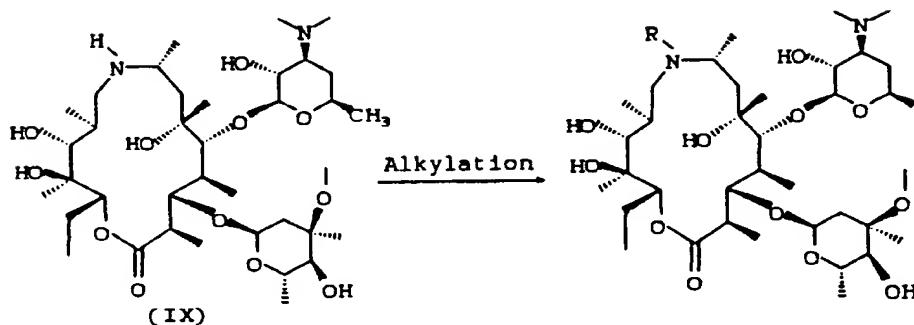
50

55

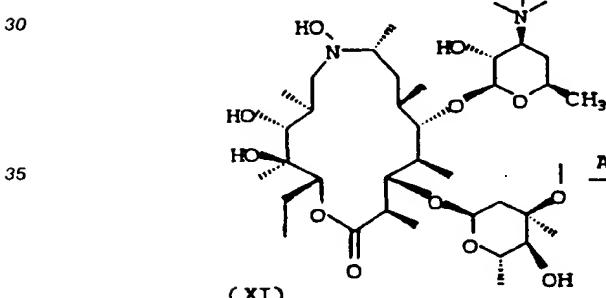
FLOW CHART (Cont'd)

5 (VII) or (VIII)

10 Reduction



25 Oxidation

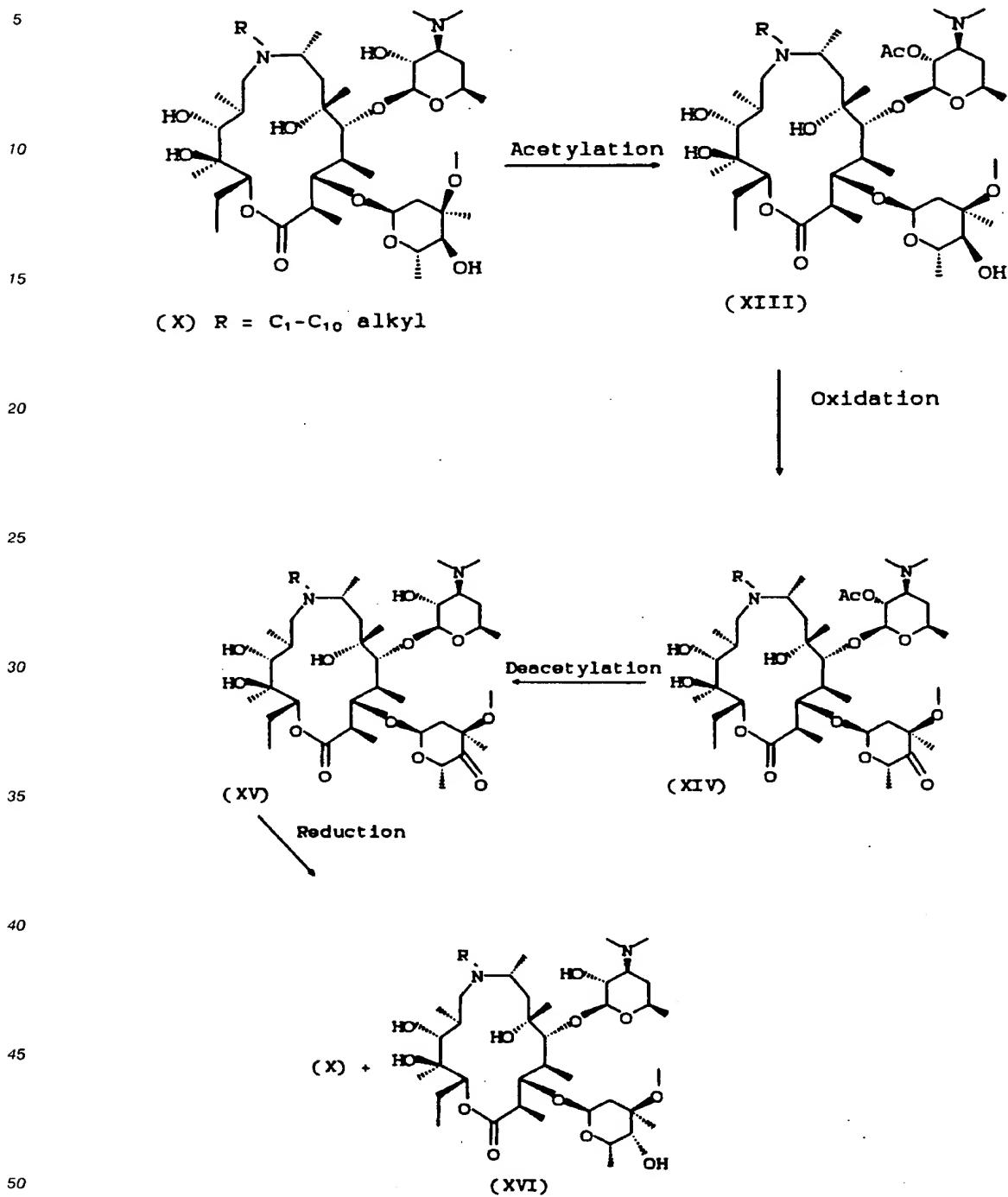


Deoxygenation

45

50

55

FLOW CHART II

FLOW CHART III

5

10

15

20

25

30

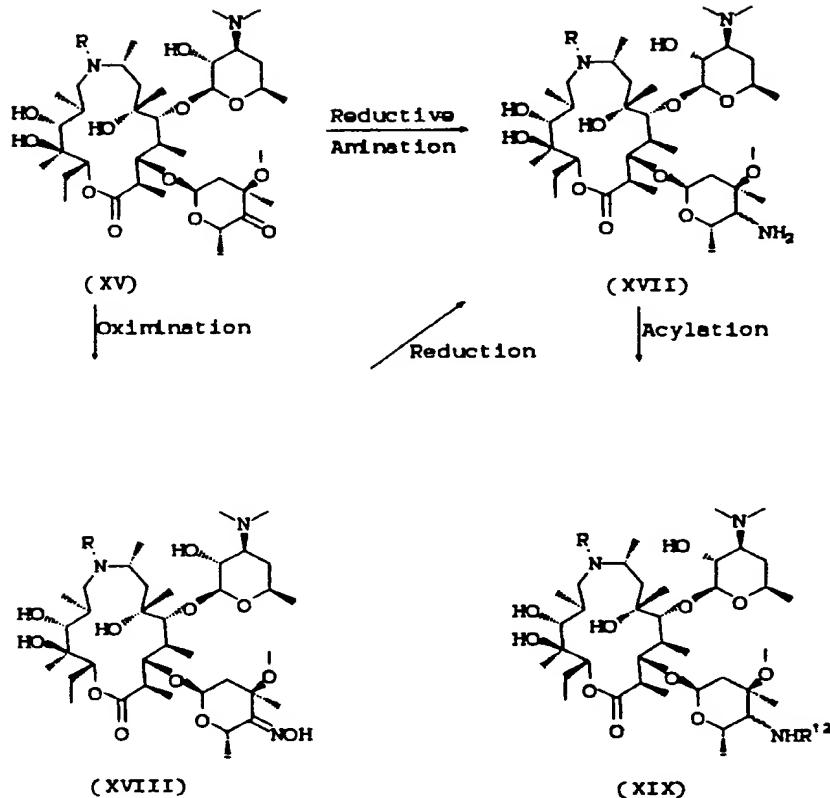
35

40

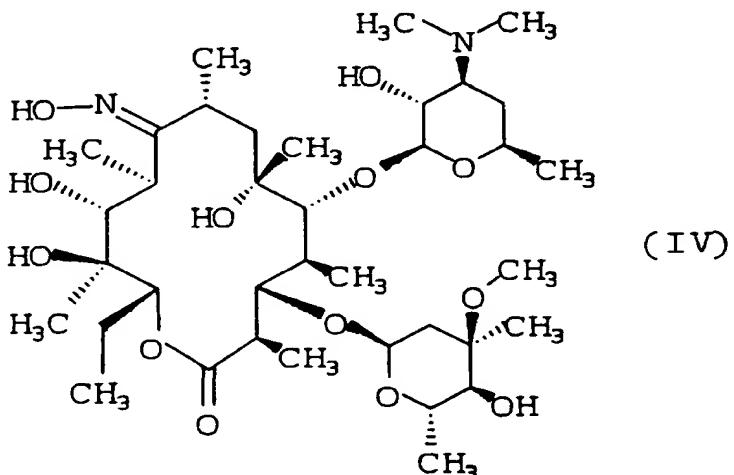
45

50

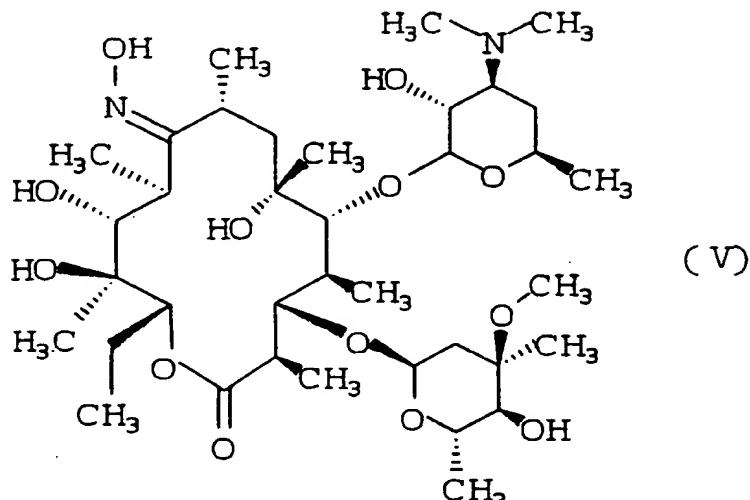
55

Isomerization of (9E)-9-Deoxy-9-hydroxyiminoerythromycin A to the (9Z) isomer

In a single step procedure, (9Z)-9-deoxy-9-hydroxyimininoerythromycin A of the structure:



is obtained by reacting (9E)-9-deoxo-9-hydroxyiminoerythromycin A of the structure:



with a base in the presence of a protic or an aprotic solvent. Preferably, the base is an alkali metal hydroxide and the solvent is an alcohol. Most preferably, the base is lithium hydroxide (as the monohydrate) and the solvent is ethanol.

25 Optimization of the method of the isomerization step requires a ratio of the aforementioned base sufficient to substantially deprotonate the hydroxyimino group of the (E) isomer. Furthermore, the oxime anion must be reasonably stable under the reaction conditions for the time period required to complete the isomerization process.

30 Upon addition of the base to the (E) isomer, an equilibrium condition is created as shown in the following equation:

35

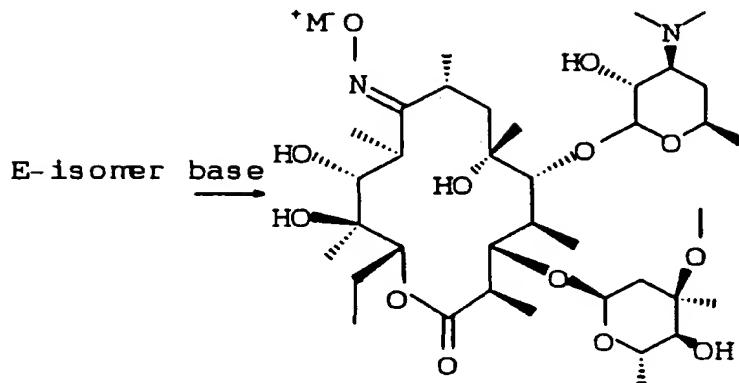
40

45

50

55

5



10

15

20

25

30

35

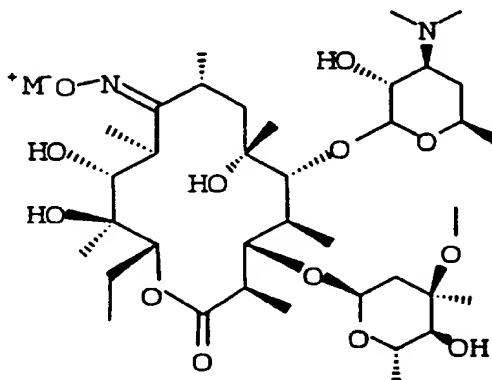
40

45

workup

E-oxime anion

E-oxime

+  
Z-oximewhere  $^+M$   
is a suitable  
counterion

Z-oxime anion

The workup performed on the anions includes protonation of the oxime anions to give the neutral oxime product mixture from which the desired (Z)-isomer is isolated by crystallization or by chromatography followed by crystallization.

The relative amounts of (E) and (Z) oxime anions (and neutral oximes after the workup) in the equilibrium mixture can be controlled and depends on a number of factors. These include: (a) the strength and quantity of the base reagent, (b) the size and polarizability of the counterion  $M^+$ , and (c) the reaction solvent.

Suitable bases include hydroxides, alkoxides, carbonates, amides, amines and hydrides.

The following list of reagents is given to illustrate suitable bases and solvents, although the list is not to be taken as exhaustive and other bases and solvents known to those of ordinary skill in the art are not excluded. Preferred bases and solvents are indicated by one asterisk and most preferred bases are indicated by a dagger.

Bases1. Hydroxides

5	* † LiOH	lithium hydroxide
	* † NaOH	sodium hydroxide
	* KOH	potassium hydroxide
	CsOH	cesium hydroxide
	Ca(OH) <sub>2</sub>	calcium hydroxide
10	Mg(OH) <sub>2</sub>	magnesium hydroxide
	* Me <sub>4</sub> NOH	tetramethylammonium hydroxide
	BnMe <sub>3</sub> NOH	benzyltrimethylammonium hydroxide
	Et <sub>4</sub> NOH	tetraethylammonium hydroxide
	Bu <sub>4</sub> NOH	tetrabutylammonium hydroxide

15 2. Alkoxides

20	* † LiOMe	lithium methoxide
	* † LiOEt	lithium ethoxide
	LiOiPr	lithium isopropoxide
	LiOnBu	lithium n-butoxide
	LiOsBu	lithium sec-butoxide
	* † NaOMe	sodium methoxide
	* † NaOEt	sodium ethoxide
25	NaOPr	sodium n-propoxide
	NaOiPr	sodium iso-propoxide
	NaOnBu	sodium n-butoxide
	NaOsBu	sodium sec-butoxide
	NaOtBu	sodium tert-butoxide
30	NaOSiMe <sub>3</sub>	sodium trimethylsilanoate
	KOMe	potassium methoxide
	* KOEt	potassium ethoxide
	KOtBu	potassium tert-butoxide
	KOSiMe <sub>3</sub>	potassium trimethylsilanoate
35	KOsBu	potassium sec-butoxide
	CsOtBu	cesium tert-butoxide
	Ca(OMe) <sub>2</sub>	calcium methoxide
	* Mg(OEt) <sub>2</sub>	magnesium ethoxide
	Ti(OEt) <sub>4</sub>	titanium (IV) ethoxide
40	Ti(OiPr) <sub>4</sub>	titanium (IV) isopropoxide
	BnMe <sub>3</sub> NOMe	benzyltrimethylammoniummethoxide

3. Carbonates

45	K <sub>2</sub> CO <sub>3</sub>	potassium carbonate
	* Cs <sub>2</sub> CO <sub>3</sub>	cesium carbonate
	Na <sub>2</sub> CO <sub>3</sub>	sodium carbonate

4. Amides (for use in aprotic solvents)

50	LiNH <sub>2</sub>	lithium amide
	LiNMe <sub>2</sub>	lithium dimethylamide
	* LiNiPr <sub>2</sub>	lithium diisopropylamide
	LiN(C <sub>6</sub> H <sub>11</sub> ) <sub>2</sub>	lithium dicyclohexylamide
55	LiN(SiMe <sub>3</sub> ) <sub>2</sub>	lithium bis(trimethylsilyl) amide
	NaNH <sub>2</sub>	sodium amide
	KN(SiMe <sub>3</sub> ) <sub>2</sub>	potassium bis(trimethylsilyl) amide

5. Amines

5	* TMG	1,1,3,3-tetramethyl guanidine
	DBU	1,8-diazabicyclo[5.4.0] undec-7-ene
	proton sponge	1,8-bis(dimethylamino)-naphthalene

6. Hydrides (for use in aprotic solvents)

10	LiH	lithium hydride
	* NaH	sodium hydride
	KH	potassium hydride

7. Solvents15 a. Protic

20	H <sub>2</sub> O	(generally used in combination with an alcohol solvent)
	* † MeOH	methanol
	* † EtOH	ethanol
	* iPrOH	isopropanol
	n-BuOH	normal-butanol
	s-BuOH	sec-butanol
	t-BuOH	tert-butanol

25 b. Aprotici. Nonpolar (as a group, these are generally used in combination with a protic or polar solvent)

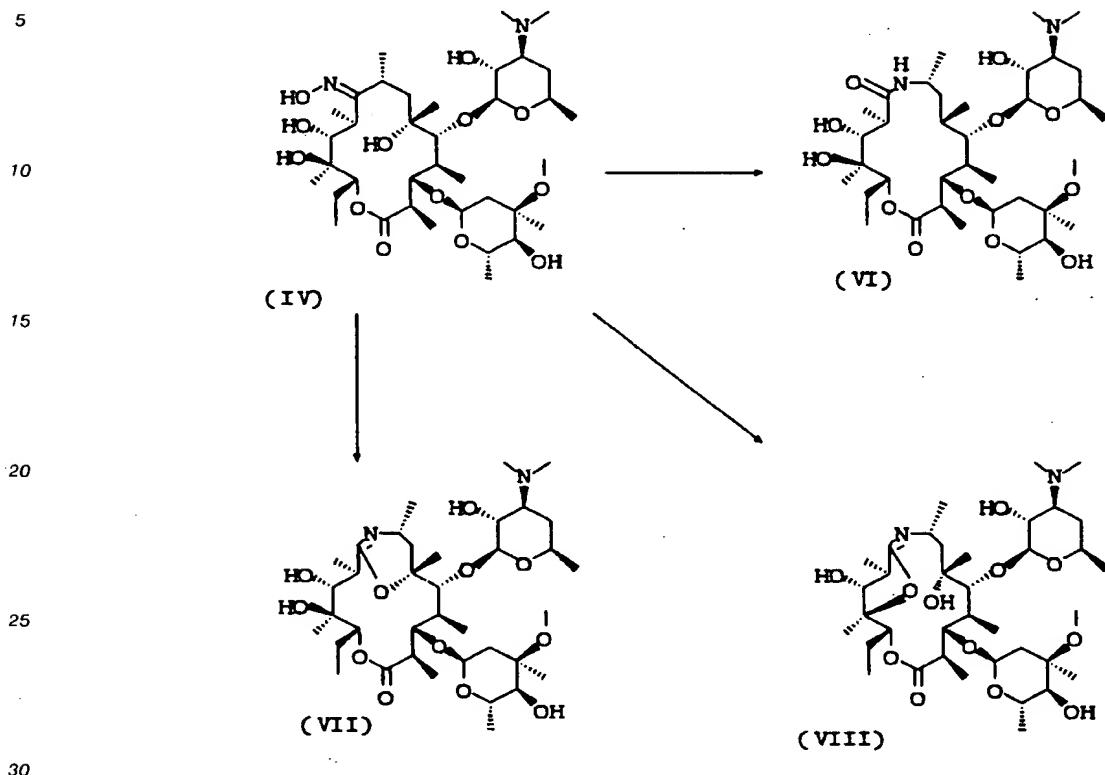
30	Et <sub>2</sub> O	diethyl ether
	THF	tetrahydrofuran
	DME	dimethoxyethane
	PhMe	toluene
	CH <sub>2</sub> Cl <sub>2</sub>	dichloromethane
	CHCl <sub>3</sub>	chloroform

35 ii. Polar

40	* DMF	dimethylformamide
	DMAC	dimethylacetamide
	DMI	1,3-dimethyl-2-imidazolidinone
	NEP	1-ethyl-2-pyrrolidinone
	* NMP	1-methyl-2-pyrrolidinone
	HMPA	hexamethylphosphoramide
	MeNO <sub>2</sub>	nitromethane
45	* MeCN	acetonitrile
	dioxane	
	pyridine	
	* DMSO	dimethylsulfoxide

Preferably, the isomerization reaction is carried out at a concentration of 1-25% w/v of E-oxime to solvent, and most preferably at 10% w/v. The amount of base used is preferably 1.0-10.0 molar equivalents of base, more preferably 1.0-3.0 molar equivalents, and most preferably 2.0 molar equivalents. The reaction is generally run at a temperature of from 0 °C to 80 °C, and more preferably at 22-25 °C. The reaction can be allowed to run for 0.5 hours to 20 days, but most preferably is carried out over 20-24 hours.

## Beckmann Rearrangement of (9Z)-9-Deoxy-9-hydroxyiminoerythromycin A



The conversion of (9Z)-9-deoxy-9-hydroxyiminoerythromycin A (IV) to the 8a-aza-8a-homoerythromycin products (VI), (VII) and (VIII) is accomplished by means of the Beckmann rearrangement (see "Comprehensive Organic Chemistry", I. O. Sutherland (Ed.), Pergamon Press, New York, 1979, Vol. 2, pgs. 398-400 and 967-968). In general, the Beckmann rearrangement of ketoximes leads to carboxamides and, in cyclic systems, to ring expanded lactams. The mechanism of the rearrangement involves initial conversion of the oxime hydroxyl group to a leaving group which is then lost with concomitant migration of the oxime carbon substituent that is situated *anti* to the leaving group. In aqueous media, the intermediate nitrilium cation thus formed usually reacts with water to afford the amide product. The nitrilium intermediate can also be trapped by other suitable nucleophiles thereby leading to imino products such as imidates and amidines.

The Beckmann rearrangement has been accomplished under a variety of acidic, neutral and basic conditions. Common acidic reagents that promote the transformation include concentrated sulfuric acid, polyphosphoric acid, thionyl chloride, phosphorous pentachloride, sulfur dioxide, and formic acid. These reagents are generally not applicable to the rearrangement of oxime (IV) due to the sensitivity of the macrolide molecule, and especially the cladinose sugar residue, to acidic conditions. Efficient Beckmann rearrangement also occurs by heating the oxime with silica gel in xylene or under mildly basic conditions by heating the oxime in hexamethylphosphoramide. These conditions are not particularly valuable for the conversion of (IV) to products (VI), (VII) and (VIII) due to competing isomerization of the oxime function under the reaction conditions.

A preferred method for effecting the Beckmann rearrangement involves initial O-sulfonylation of the oxime group with an alkylsulfonyl halide, arylsulfonyl halide or arylsulfonic anhydride. The intermediate oxime sulfonate thus formed can be isolated or, as more commonly practiced, converted *in situ* to the rearranged products. The sulfonation and rearrangement reactions are generally performed in the presence of an organic or inorganic base. This method is particularly valuable for the conversion of oxime (IV) to the rearranged products (VII), (VIII), and (VIII).

Preferred sulfonating reagents for effecting the rearrangement of oxime (IV) include methanesulfonyl chloride, benzenesulfonyl chloride, 4-acetamidobenzenesulfonyl chloride, p-toluenesulfonyl chloride, ben-

zenesulfonic anhydride, and p-toluenesulfonic anhydride. The reaction is carried out in the presence of an inorganic base such as sodium bicarbonate or potassium carbonate, or in the presence of an organic base such as pyridine, 4-dimethylaminopyridine, triethylamine, or N,N-disopropylethylamine. Suitable solvents include aqueous mixtures such as aqueous acetone or aqueous dioxane and organic solvents such as dichloromethane, chloroform, ethyl acetate, diethyl ether, tetrahydrofuran, toluene, acetonitrile, and pyridine. Mixtures of organic solvents, especially those containing pyridine, are highly useful. The reaction is generally performed using 1-3 molar equivalents of the sulfonating agent and one or more molar equivalents of base at a reaction temperature of -20 °C to 50 °C. Pyridine is often used as both solvent and base.

The distribution of products resulting from Beckmann rearrangement of oxime (IV) depends on the particular reaction conditions employed. For example, when the rearrangement is effected with p-toluenesulfonyl chloride and sodium bicarbonate in aqueous acetone, the major products are the lactam (VI) and the 6,9-bridged iminoether (VII). When the reaction is conducted under anhydrous conditions such as p-toluenesulfonyl chloride in pyridine, the major products are the 6,9-bridged and 9,12-bridged iminoethers (VII) and (VIII). The ratio of products (VII) and (VIII) is also effected by the addition of cosolvents, by temperature, and by the initial oxime concentration. In general, increasing the proportion of pyridine as solvent, increasing the reaction temperature, and decreasing the initial oxime concentration all favor the formation of the 9,12-product (VIII) over that of the 6,9-product (VII).

A particularly preferred method for effecting the Beckmann rearrangement of oxime (IV) involves the addition of a solution of 2.5 molar equivalents of p-toluenesulfonyl chloride in diethyl ether to a solution of the oxime in pyridine at 0-5 °C. Oxime O-sulfonylation and subsequent rearrangement occur under the reaction conditions to afford a 1:3 mixture of iminoether products (VII) and (VIII).

The products of the Beckmann rearrangement of oxime (IV) are conveniently purified by chromatographic methods. For example, the lactam (VI) is easily separated from iminoether (VII) using column chromatography on silica gel or by reverse phase, high-pressure liquid chromatography. Products (VII) and (VIII) can also be separated by chromatographic methods, and the (VIII) thus obtained can be further purified by crystallization from nitromethane. However, it is generally expedient to simply carry the mixture of (VII) and (VIII) isomers into the subsequent reduction step without further purification.

As previously noted, Beckmann rearrangement of oxime (IV) under anhydrous conditions leads to a product mixture comprised of the 6,9- and 9,12-bridged iminoethers (VII) and (VIII). The 9,12-bridged product, which is formed by stereoselective internal trapping of the intermediate nitrilium species by the hydroxyl group at C-12, is initially isolated as a mixture of major and minor forms that are isomeric about the imino double bond. The initial mixture of isomers equilibrates at room temperature, both in solution or on storing as a crude foam, to approximately a 1:1 mixture of isomers. The first-formed, major isomer can be isolated from the mixture by crystallization from nitromethane solution. However, it should be noted that the equilibration of the 9,12-bridged iminoethers (VIII) is of no consequence to the overall process since both forms are easily reduced in the next step to 9-deoxo-8a-aza-8a-homoerythromycin A.

40

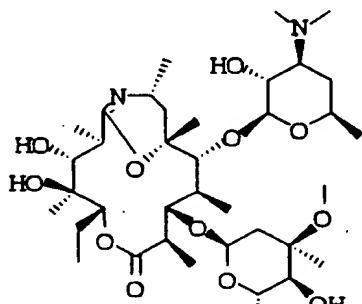
45

50

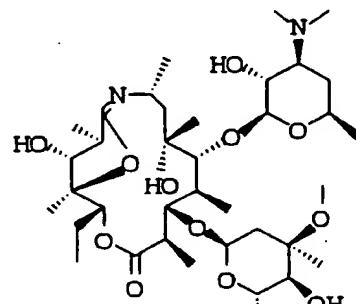
55

Reduction of 9-Deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (VII) and 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (VIII)

5



10



15

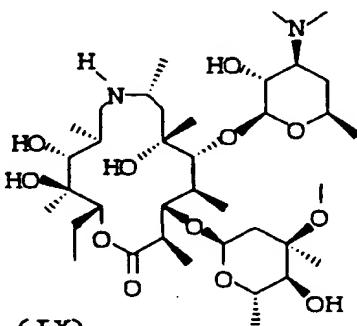
(VII)

(VIII)

20



25



30

(IX)

35

Compounds (VII) and (VIII) can be viewed as cyclic imides (or imidic acid esters) of the macrocyclic lactam (VI). The imides are formally derived from compound (VI) by transannular addition of the hydroxy groups at positions 6 and 12 to the lactam carbonyl group, followed by elimination of water. However, 40 imides (VII) and (VIII) most likely arise by intramolecular interception of the Beckmann rearrangement nitrilium intermediate with the 6- and 12-hydroxy groups. In structure (VII), the imide function (-N=C-O-) lies completely within a 6-membered ring thereby giving rise to a 5,6-dihydro-1,3-oxazine system. By contrast, the second structure (VIII) has the imino nitrogen atom exocyclic to a 5-membered ring containing the oxygen atom thereby giving rise to a 2-iminotetrahydrofuran system.

45 A number of reagents are known for the reduction of imides to the corresponding amines (see "The Chemistry of Amidines and Imides", S. Patai (Ed.), John Wiley and Sons, 1975, pgs. 460-461 and "Comprehensive Organic Chemistry", I. O. Sutherland (Ed.), Pergamon Press, New York, 1979, Vol. 2, pg. 495). These include metal/proton-donor combinations such as sodium amalgam in acid solution or sodium in ethanol or liquid ammonia, catalytic hydrogenation under pressure, and complex metal hydrides such as 50 lithium aluminum hydride and sodium borohydride. The electrochemical reduction of imides is also reported to give amines in good yield.

55 A method of choice for reducing the macrocyclic imides (VII) and (VIII) to the amine (IX) uses a complex metal hydride in an appropriate solvent system. Suitable hydride reagents include lithium aluminum hydride; sodium borohydride, sodium cyanoborohydride, and diisobutyl aluminum hydride. Both lithium aluminum hydride and diisobutyl aluminum hydride require the use of anhydrous solvents such as benzene, toluene, diethyl ether, tetrahydrofuran and dimethoxyethane, whereas sodium borohydride and sodium cyanoborohydride can be used in the presence of water or in alcoholic solvents such as methanol, ethanol, isopropanol, and ethylene glycol. When sodium cyanoborohydride is used the reaction medium is

usually acidified by addition of aqueous acid ( $\text{pH} \geq 3$ ) or acetic acid. The reaction is generally accomplished by treating the imidate with 1-5 molar equivalents of reductant for 1-20 hours at a temperature ranging from -20 °C to 50 °C.

5 A particularly preferred method for reducing imidates (VII) and (VIII) to the amine (IX) employs 2-3 molar equivalents of sodium borohydride in methanol or ethylene glycol at a temperature of 0 °C to 25 °C. Ethylene glycol serves the dual purposes of activating the borohydride agent and of breaking up borate ester complexes of the amine product.

10 A second method of choice for effecting the reduction of imidates (VII) and (VIII) to the amine (IX) is catalytic hydrogenation at high pressure. The reaction is usually accomplished by shaking a mixture of imidate and catalyst in a suitable solvent such as methanol, ethanol, aqueous dioxane or acetic acid at a hydrogen pressure of 1000-3000 psi for 2-20 hours at ambient temperature. Suitable catalysts include noble metals and their oxidized forms such as platinum on carbon, platinum oxide (Adams' catalyst), palladium on carbon, palladium hydroxide on carbon (Pearlman's catalyst) and rhodium on carbon. An especially preferred method for reducing imidate (VII) uses nearly an equivalent weight of platinum oxide catalyst in 15 acetic acid at 2000 psi hydrogen for 18-20 hours at room temperature.

#### Methylation of 9-Deoxo-8a-aza-8a-homoerythromycin A

20 Secondary amines such as (IX) can be reductively methylated to tertiary amines using formaldehyde in the presence of a reducing agent. Suitable reductants for this reaction include hydrogen in the presence of a noble metal catalyst, Raney nickel, sodium borohydride, sodium cyanoborohydride, and formic acid. The reaction can be conducted in a number of organic solvents, for example methanol, ethanol, acetonitrile, chloroform, tetrahydrofuran or dioxane, with or without the presence of added water. Perhaps the most common of these methylation procedures is the Eschweiler-Clarke method, which involves the reaction of 25 the amine with formaldehyde in the presence of formic acid.

30 Application of the Eschweiler-Clarke procedure to compound (IX) affords the ring methylated product (X) wherein R is  $\text{CH}_3$ . The reaction is accomplished by treating (IX) with 1-2 molar equivalents of formaldehyde and 2-3 molar equivalents of formic acid in an inert solvent at 20-100 °C. A preferred system uses 37% aqueous formaldehyde and formic acid in carbon tetrachloride or chloroform heated at reflux for 1-2 days. The product is conveniently isolated by crystallization from aqueous ethanol.

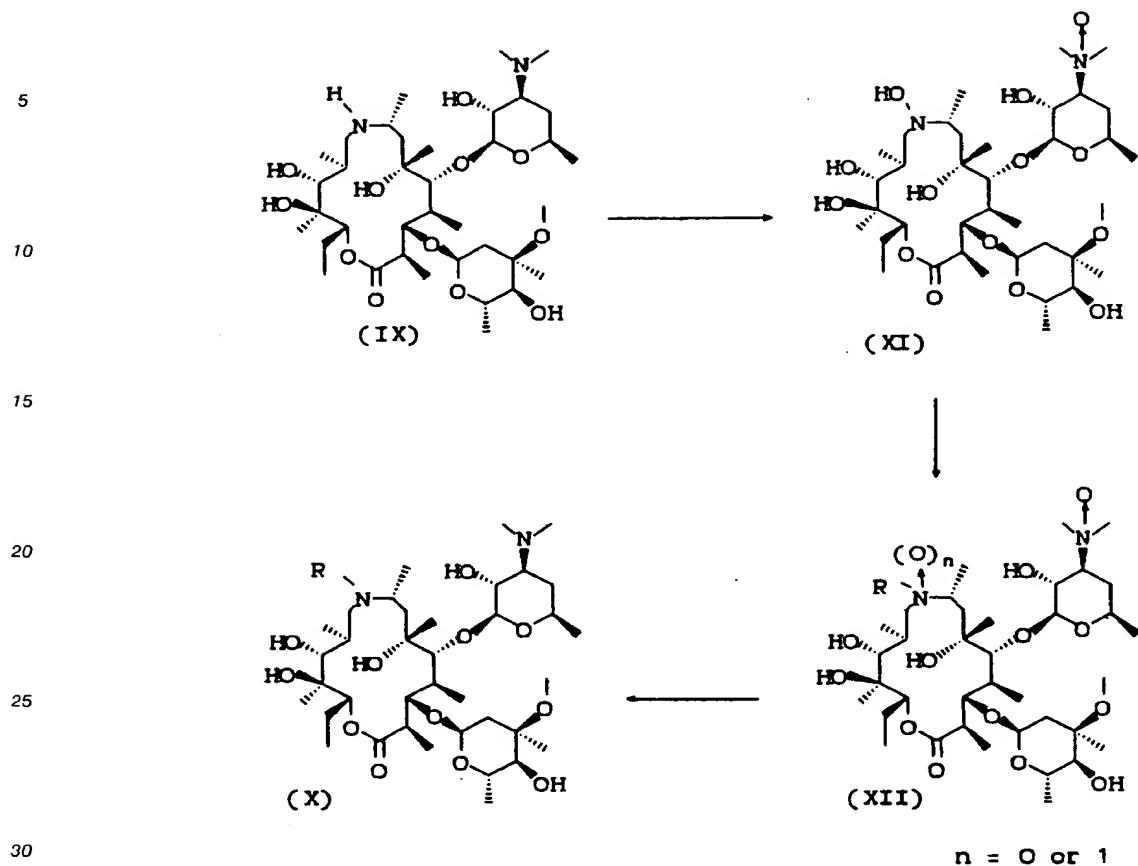
35

40

45

50

55



The methylation of compound (IX) can also be accomplished using a three-step procedure (see G. M. Bright, et al., *J. Antibiotics*, **41**, 1029 (1988) and U.S. Patent No. 4,474,768) in which (IX) is first oxidized to the N-hydroxide N'-oxide intermediate (XI), then treated with a methylating agent to afford the intermediate product (XII), and finally deoxygenated to the desired product. In this approach, the N'-oxygen serves as a temporary protecting group to prevent quaternization at the desosamine dimethylamino group.

The oxidation step is conducted in an inert solvent using hydrogen peroxide or a peracid such as peracetic acid or 3-chloroperoxybenzoic acid as the oxidant. Suitable solvents for the reaction include dichloromethane, chloroform, tetrahydrofuran, dioxane, methanol, ethanol and acetic acid. In general, a water miscible solvent such as methanol or acetic acid is used with the water-soluble oxidants hydrogen peroxide and peracetic acid, whereas an anhydrous solvent such as dichloromethane or tetrahydrofuran is used with 3-chloroperoxybenzoic acid. The reaction is usually accomplished with an excess of oxidant (2-40 molar equivalents) at a temperature of from 0 °C to 50 °C for up to 24 hours. A particularly preferred embodiment employs excess 30% aqueous hydrogen peroxide as oxidant in methanol solvent at room temperature for 18-20 hours.

Introduction of the ring N-methyl group is accomplished by treating the N-hydroxy-N'-oxide intermediate (XI) with a methylating agent in an inert solvent in the presence of an acid acceptor. An inert solvent is defined as one that will not react with the methylating reagent under the reaction conditions. Suitable solvents include but are not limited to dichloromethane, chloroform, tetrahydrofuran, dimethoxyethane, dimethylsulfoxide, and toluene. Of the numerous methylating agents that are known to effect alkylation at nitrogen, methyl iodide, methyl bromide, dimethyl sulfate and methyl trifluoromethanesulfonate are well suited for the present application. The acid acceptor component, which serves to neutralize the acid formed on reaction of the methylating agent with the ring nitrogen atom, can be an inorganic base such as an alkali metal hydroxide or carbonate, or a hindered amine base. Examples of suitable acid acceptors are sodium bicarbonate, potassium carbonate, potassium hydroxide, and 2,6-lutidine. The methylation reaction is generally accomplished using a large excess (10-75 molar equivalents) of both the methylating agent and the acid acceptor at a temperature of 0 °C-80 °C for 1-20 hours. A preferred method involves stirring

compound (XII) with approximately 40 molar equivalents of methyl iodide and 70 molar equivalents of anhydrous potassium carbonate in dichloromethane at room temperature. The product of the methylation reaction is usually obtained as a mixture of components (XII, n = 0 and 1) wherein the ring nitrogen atom has suffered partial deoxygenation. These components can be separated by chromatography, but are generally used without purification in the following deoxygenation step.

The final step of the sequence, the deoxygenation reaction of (XII) to provide (X), is readily accomplished by catalytic hydrogenation. The hydrogenation reaction is carried out at a temperature of 18°C to 25°C and at hydrogen pressures of from 15 psi to 2000 psi in an inert solvent. Suitable catalysts are noble metals and their oxides such as palladium on carbon, palladium hydroxide on carbon, platinum oxide, platinum on carbon, and rhodium on carbon. Representative inert solvents for the catalytic reduction are methanol, ethanol, tetrahydrofuran, dioxane, acetic acid and mixtures thereof. A typical catalytic reduction procedure uses ethanol as solvent, a hydrogen pressure of 45 psi, and 10% palladium on carbon as catalyst at a substrate to catalyst ratio of 1:1 to 1:2.

The reductive deoxygenation of (XII) to (X) can also be accomplished with a number of chemical reductants. Representative reagents of this type include metal hydrides such as sodium borohydride or sodium cyanoborohydride, zinc in acetic acid, and triphenylphosphine.

In the foregoing discussion, the N-alkylation of 9-deoxo-8a-aza-8a-homoerythromycin A (IX) was illustrated for the case wherein the alkyl group (R) is a methyl group. Other alkyl groups are introduced using similar methodology well known to those of ordinary skill in the art. For example, reductive alkylation of IX with a variety of C<sub>2</sub>-C<sub>10</sub> carboxaldehydes, instead of formaldehyde, provides the higher homologs (X) wherein R is C<sub>2</sub>-C<sub>10</sub> alkyl. Alternatively, the N-hydroxy-N<sup>1</sup>-oxide intermediate (XI) reacts with a variety of C<sub>2</sub>-C<sub>10</sub> alkylating agents to afford compounds XII wherein the ring nitrogen is substituted with a C<sub>2</sub>-C<sub>10</sub> alkyl group. Deoxygenation provides the N-alkyl products (X). Suitable alkylating agents for this process include, but are not limited to, C<sub>2</sub>-C<sub>10</sub> alkyl iodides, tosylates, mesylates, and triflates.

Turning now to Flow Chart II there is shown in the first step conversion of compounds of formula X via acetylation to a protected moiety. Protection of the 2'-hydroxyl group is necessary for the subsequent oxidation step and is generally achievable by use of a lower alkanoyl protecting group such as acetyl or propionyl. Typically, acetic anhydride and pyridine in dichloromethane will attach the acetyl group to the 2' hydroxyl when allowed to react at room temperature for several hours.

Next, the 2'-protected intermediate is oxidized at the 4"-hydroxyl substituent to the corresponding 4"-oxo (keto) compound, via treatment with dimethyl sulfoxide and a suitable activating agent in the presence of a base. Typical reaction conditions for the oxidation are either: (a) Moffatt oxidation which uses N-ethyl-N'-(N,N-dimethylaminopropyl)carbodiimide and dimethylsulfoxide in the presence of pyridinium trifluoroacetate; or (b) the Swern oxidation in which oxalyl chloride and dimethylsulfoxide in dichloromethane is followed by the addition of triethylamine or alternatively trifluoroacetic anhydride and dimethylsulfoxide in dichloromethane is followed by the addition of triethylamine.

In the next step, removal of the 2'-protecting group is achieved by solvolysis with methanol at room temperature for 1 to 2 days, followed by *in vacuo* evaporation of the methanol.

The subsequent reduction step reduces the 4"-oxo group to the 4"-epi hydroxy isomer of (II) via catalytic hydrogenation, as with Raney nickel and hydrogen. Also, a complex metal hydride such as sodium borohydride in methanol can be used to achieve reduction. To the extent isomeric mixtures at the 4" position are formed, they are conveniently separated by silica gel column chromatography or by HPLC methods.

Turning now to Flow Chart III, there is shown the reductive amination that directly converts the 4" oxo to the corresponding 4"-amino. This is accomplished by either: (a) sodium cyanoborohydride and ammonium acetate in methanolic acetic acid; or (b) catalytic hydrogenation in the presence of an ammonia source, such as hydrogen and ammonium acetate in methanol with Raney nickel or 10% palladium on carbon. The resulting amino isomers at the 4" position can be separated by HPLC or by chromatography on silica gel or alumina.

Alternatively, the 4"-amino derivative can be prepared from the 4"-oxo via intermediate oximation to the corresponding 4" oxime followed by reduction to the 4"-amine. Oximation is achieved by use of hydroxylamine hydrochloride in pyridine or hydroxylamine hydrochloride and a base such as triethylamine or imidazole in methanol. Reduction of the oxime is accomplished by either: (a) catalytic hydrogenation with hydrogen and Raney nickel in ethanol, or (b) reduction with titanium trichloride in ammonium acetate buffered methanol containing sodium cyanoborohydride.

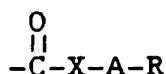
The 4"-amine can be acylated or sulfonylated to the corresponding amide, carbamate suitable reagents for this process include, but are not limited to: (a) R"COCl (R"CO)<sub>2</sub>O, R"SO<sub>2</sub>Cl, or (R"SO<sub>2</sub>)<sub>2</sub>O in dichloromethane containing triethylamine, where R" is any substituent of interest; (b) R"CO<sub>2</sub>H and

dicyclohexylcarbodiimide in dichloromethane; or (c) R"CO<sub>2</sub>H and iso-butylchloroformate in dichloromethane containing N-methylmorpholine.

Structural formula (XIX) shows an N-acyl group, R<sup>12</sup>. R<sup>12</sup> is preferably either a substituted or unsubstituted, C<sub>1</sub>-C<sub>10</sub> alkylcarbonyl, C<sub>1</sub>-C<sub>10</sub> alkoxy carbonyl, arylcarbonyl, aryl C<sub>1</sub>-C<sub>10</sub> alkylcarbonyl, aryl C<sub>1</sub>-C<sub>10</sub> alkoxy carbonyl, or aryl or heteroaryl-carbonyl group, or an arylsulfonyl group. These substituents can be 1-3 fluoro, aryl, amino, substituted amino wherein said substituent is formyl or C<sub>1</sub>-C<sub>10</sub> alkylcarbonyl; cyano, isonitrilo, carboxy, carbamoyl, Br, Cl, disubstituted amino wherein said substituents are C<sub>1</sub>-C<sub>10</sub> alkyl; nitro, heterocyclyl, heteroaryl, substituted carbamoyl wherein the nitrogen atom is mono or di-substituted with C<sub>1</sub>-C<sub>10</sub> allyl; aryloxy, arythio, C<sub>1</sub>-C<sub>10</sub> alkoxy carbonyl hydroxy, C<sub>1</sub>-C<sub>10</sub> alkoxy, mercapto, C<sub>1</sub>-C<sub>10</sub> alkylthio, C<sub>1</sub>-C<sub>10</sub> alkylsulfinyl, C<sub>1</sub>-C<sub>10</sub> alkylsulfonyl, unsubstituted or substituted iminomethyl wherein said substituent is C<sub>1</sub>-C<sub>10</sub> alkyl, sulfamoyl, unsubstituted or substituted C<sub>1</sub>-C<sub>10</sub> alkylamino wherein said substituent is formyl; and C<sub>1</sub>-C<sub>10</sub> alkylcarbonyl.

Also preferably, R<sup>12</sup> is hydrogen, arylsulfonyl, halogen-substituted arylsulfonyl, thienylsulfonyl, furanyl sulfonyl, or

15



20

where

X is a connecting bond, O or NH;

A is a connecting bond, or C<sub>1</sub>-C<sub>3</sub> alkylene;

R is hydrogen, C<sub>1</sub>-C<sub>10</sub> alkyl, aryl, heteroaryl, aralkyl, heterocyclyl, or C<sub>3</sub>-C<sub>7</sub> cycloalkyl, any of which R

25 groups other than hydrogen can be substituted by one or more of halogen, hydroxyl, C<sub>1</sub>-C<sub>3</sub> alkoxy, cyano, nitro, amino, mono- or di-(C<sub>1</sub>-C<sub>3</sub>) alkylamino, mercapto, C<sub>1</sub>-C<sub>3</sub> alkylthio, C<sub>1</sub>-C<sub>3</sub> alkylsulfinyl, C<sub>1</sub>-C<sub>3</sub> alkylsulfonyl, arylthio, arylsulfinyl, sulfamoyl, arylsulfonyl, carboxy, carbamoyl, C<sub>1</sub>-C<sub>3</sub> alkylcarbonyl, C<sub>1</sub>-C<sub>3</sub> alkoxycarbonyl, or C<sub>1</sub>-C<sub>3</sub>-substituted halogen.

As antibiotics, the compounds of formula (II) can be administered in such oral dosage forms as tablets, 30 capsules, pills, powders, granules, elixirs, tinctures, suspensions, syrups and emulsions. Likewise, they may also be administered in intravenous, intraperitoneal, subcutaneous or intramuscular form, all using forms well known to those of ordinary skill in the pharmaceutical arts. In general, the preferred form of administration is oral. An effective but non-toxic amount of the compound can be employed as a mammalian antibiotic.

35 The dosage regimen utilizing the compounds of formula (II) is selected in accordance with a variety of factors including type, species, age, weight, sex and medical condition of the patient; the severity of the condition to be treated; the route of administration; the renal and hepatic function of the patient; and the particular compound or salt thereof employed. An ordinarily skilled physician or veterinarian can readily determine and prescribe the effective amount of the drug required to prevent, counter or arrest the progress 40 of the condition.

45 Dosages of the compounds of formula (II), when used for the indicated effects, will range between about 0.2 mg per kg of body weight per day (mg/kg/day) to about 120 mg/kg/day and preferably 4-50 mg/kg/day. Advantageously, the compound may be administered in a single daily dose, or the total daily dosage may be administered in divided doses of two, three or four times daily.

Furthermore, the compounds of formula (II) can be administered in topical, otic or ophthalmic form via use of suitable vehicles.

In the methods of using the compounds (II), they can form the active ingredient, and are typically administered in admixture with suitable pharmaceutical diluents, excipients or carriers (collectively referred to herein as "carrier" materials) suitably selected with respect to the intended form of administration, that is, 50 oral tablets, capsules, elixirs, syrups, and the like, and consistent with conventional pharmaceutical practices.

For instance, for oral administration in the form of a tablet or capsule, the active drug component can be combined with an oral, non-toxic, pharmaceutically acceptable, inert carrier such as lactose, starch, sucrose, glucose, methyl cellulose, magnesium stearate, dicalcium phosphate, calcium sulfate, mannitol, 55 sorbitol, and the like; for oral administration in liquid form, the oral drug components can be combined with any oral, non-toxic, pharmaceutically acceptable inert carrier such as ethanol, glycerol, water, and the like. Moreover, when desired or necessary, suitable binders, lubricants, disintegrating agents and coloring agents can also be incorporated into the mixture. Suitable binders include starch, gelatin, natural sugars such as

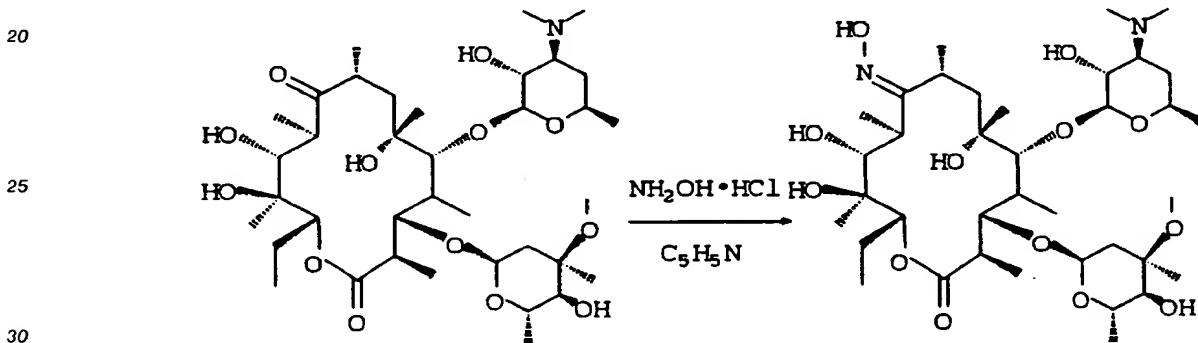
glucose or beta-lactose, corn sweeteners, natural and synthetic gums such as acacia, tragacanth or sodium alginate, carboxymethylcellulose, polyethylene glycol, waxes, and the like. Disintegrators include, without limitation, starch, methyl cellulose, agar, bentonite, xanthan gum, and the like.

The compounds of formula (II) can also be administered in the form of liposome delivery systems, such as small unilamellar vesicles, large unilamellar vesicles and multilamellar vesicles. Liposomes can be formed from a variety of phospholipids, such as cholesterol, stearylamine or phosphatidylcholines.

The compounds of formula (II) may also be coupled with soluble polymers as targetable drug carriers. Such polymers can include polyvinylpyrrolidone, pyran copolymer, polyhydroxypropylmethacrylamide phenyl, polyhydroxyethylaspartamide-phenol, or polyethyleneoxide-polylysine substituted with palmitoyl residues. Furthermore, the compound of formula (II) may be coupled to a class of biodegradable polymers useful in achieving controlled release of a drug, for example, polylactic acid, polyglycolic acid, copolymers of polylactic and polyglycolic acid, polyepsilon caprolactone, polyhydroxy butyric acid, polyorthoesters, polyacetals, polydihydropyrans, polycyanoacrylates and cross-linked or amphipathic block copolymers of hydrogels.

15

#### EXAMPLE 1



#### Preparation of (9E)-9-Deoxy-9-hydroxyiminoerythromycin A

Hydroxylamine hydrochloride (224 g, 3.23 mol) was added to a solution of erythromycin A (100 g, ca. 95% pure, 0.129 mol, available from Aldrich Chemical Co., Milwaukee Wisconsin) in pyridine (500 mL). The resulting mixture was stirred at room temperature for 27 hours, and then concentrated under vacuum at ca. 40 °C. The semi-solid residue was kept under high vacuum overnight, then stirred with ethanol (600 mL) for 15 minutes and filtered. The collected solids were washed with hot (50 °C) ethanol. The combined filtrate and washing was evaporated under vacuum to a pale blue foam. The foam was shaken with water (850 mL) to give a thick emulsion which was stirred rapidly at room temperature for 2.5 hours to give a filterable precipitate. The precipitate was collected, washed with water (150 mL), and dried under vacuum to give a white solid (117.7 g).

The crude oxime hydrochloride was suspended in 5% aqueous sodium bicarbonate (1000 mL) and methylene chloride (1000 mL), and the mixture was stirred while the pH was adjusted to 9.5 by addition of 5N aqueous sodium hydroxide. The layers were separated and the aqueous portion was extracted with ethyl acetate (500 mL) and ethyl ether (500 mL). The combined organic layer and extracts were dried over sodium sulfate, filtered, and evaporated under vacuum to a white solid (92.3 g). The solid was dissolved in hot ethyl acetate (250 mL), and the solution diluted with hot hexanes (400 mL) and left overnight in a refrigerator. The crystals of (9E)-9-deoxy-9-hydroxyiminoerythromycin A were collected, washed with ice-cold hexane (250 mL), and dried under vacuum to afford a white solid (88.5 g).

IR ( $\text{CH}_2\text{Cl}_2$ ) 3560, 3400 (br), 2980, 2950, 1735, 1460, 1389, 1165, 1110, 1085, 1050, and 1010  $\text{cm}^{-1}$ .  
 $^1\text{H NMR}$  ( $\text{CDCl}_3$ )  $\delta$  5.05 (dd, H-13), 4.90 (d, H-1''), 4.38 (d, H-1'), 4.01 (m, H-5''), 3.99 (d, H-3), 3.74 (m, H-8), 3.66 (s, H-11), 3.54 (d, H-5), 3.45 (m, H-5''), 3.28 (s,  $\text{OCH}_3$ ), 3.23 (dd, H-2''), 2.96 (t, H-4''), 2.87 (m, H-2), 2.64 (q, H-10), 2.43 (m, H-3'), 2.32 (d, H-2'eq), 2.27 (s,  $\text{N}(\text{CH}_3)_2$ ), 1.98 (m, H-4), 1.87 (m, H-14eq), 1.63 (m, H-4'eq), and 1.46 (s, 6- $\text{CH}_3$ ).  
 $^1\text{H NMR}$  ( $\text{CD}_3\text{OD}$ )  $\delta$  5.19 (dd, H-13), 4.48 (d, H-1''), 4.15 (dq, H-5''), 3.98 (d, H-3), 3.76 (m, H-8), 3.70 (m, H-

5'), 3.67 (s, H-11), 3.58 (d, H-5), 3.33 (s, OCH<sub>3</sub>), 3.23 (dd, H-2'), 3.01 (d, H-4''), 2.92 (m, H-2), 2.72 (m, H-10), 2.70 (m, H-3'), 2.43 (d, H-2''eq), 2.33 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.01 (m, H-4), 1.88 (m, H-14eq), 1.72 (m, H-4'eq), 1.58 (dd, H-2''ax), 1.48 (m, H-14ax), 1.45 (s, 6-CH<sub>3</sub>), 1.26 (d, 5''-CH<sub>3</sub>), 1.23 (s, 3''-CH<sub>3</sub>), 1.14 (s, 12-CH<sub>3</sub>), 1.10 (d, 4-CH<sub>3</sub>), 1.05 (d, 8-CH<sub>3</sub>), and 0.84 (t, CH<sub>2</sub>CH<sub>3</sub>).

5 <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 175.3, 171.3, 103.1, 96.3, 83.5, 80.3, 78.1, 77.1, 75.1, 74.3, 72.6, 71.2, 70.9, 68.8, 65.4, 65.3, 49.4, 44.6, 40.3, 38.8, 37.8, 35.1, 32.6, 29.2, 27.0, 25.4, 21.5, 21.3, 18.7, 18.6, 16.3, 14.3, 10.6, and 9.3.

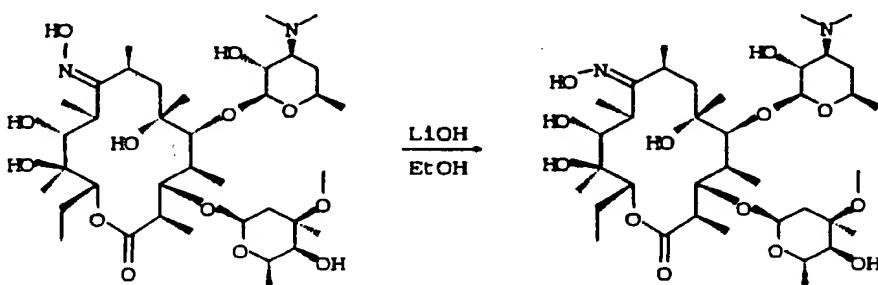
<sup>13</sup>C NMR (CD<sub>3</sub>OD) δ 177.5, 171.6, 104.0, 98.0, 84.2, 81.2, 79.3, 78.3, 76.3, 74.2, 72.9, 72.2, 69.0, 66.7, 65.2, 50.0, 46.3, 40.7, 39.3, 36.2, 32.0, 27.4, 26.7, 22.3, 22.0, 21.6, 19.3, 19.1, 17.3, 16.6, 14.8, 11.2, and 10.2.

EI Mass Spectrum, m/z 748, 590, 574, 462, 431, 416, 398, 174, 159, 158, and 116.

10

## EXAMPLE 2

15



20

### Conversion of (9E)-9-Deoxo-9-hydroxyiminoerythromycin A to (9Z)-9-Deoxo-9-hydroxyiminoerythromycin A

#### Method 1:

30

(9E)-9-Deoxo-9-hydroxyiminoerythromycin A (20.0 g, 26.7 mMol) was added to a stirred solution of lithium hydroxide monohydrate (2.25 g, 53.5 mMol) in absolute ethanol (200 mL). The solution was blanketed with nitrogen and stirred overnight at room temperature. The solvents were evaporated under vacuum and the residue was partitioned between ethyl acetate (200 mL) and brine (120 mL). The pH of the mixture was adjusted from 11 to 9.3 with 2 N hydrochloric acid. The ethyl acetate was removed and the brine was re-extracted with more ethyl acetate (2 x 200 mL). The combined ethyl acetate extracts were washed with brine (100 mL), dried with anhydrous magnesium sulfate, filtered and evaporated under vacuum to a foam (ca. 20 g).

The crude oxime mixture was dissolved in methylene chloride (220 mL) and stirred for 1 hour at room temperature to give a filterable, white solid (18.7 g). This material was dissolved in ethyl acetate (100 mL), diluted with nitromethane (100 mL), and 50 mL of solvent was evaporated under vacuum. Additional nitromethane (50 mL) was added and 80 mL of solvent was evaporated under vacuum. The solution was seeded with the (9Z)-isomer and stirred at ambient temperature for 3 hours. The resulting suspension was filtered and the solids were rinsed with nitromethane (20 mL) and dried under a stream of nitrogen to afford (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (14.8 g, 74% yield) as a white solid.

MP 157-164°C.

IR (CHCl<sub>3</sub>) 3680, 3435 (br), 2970, 2940, 1725, 1455, 1375, 1345, 1165, 1105, 1085, 1045, 1005, and 950 cm<sup>-1</sup>.

<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.01 (dd, H-13), 4.87 (d, H-1''), 4.40 (d, H-1'), 3.98 (m, H-3 and H-5''), 3.80 (s, H-11), 3.49 (m, H-5 and H-5'), 3.27 (s, OCH<sub>3</sub>), 3.21 (dd, H-2'), 2.99 (m, H-4''), 2.8 (m, H-8, H-2 and H-10), 2.74 (m, H-10), 2.43 (m, H-3'), 2.32 (d, H-2''eq), 2.27 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.91 (m, H-4), 1.87 (m, H-14eq), 1.63 (m, H-4'eq), 1.51 (m, H-2''ax and H-7), 1.42 (m, H-14ax), 1.37 (s, 6-CH<sub>3</sub>), 1.28 (d, 10-CH<sub>3</sub>), 1.24 (d, 5''-CH<sub>3</sub>), 1.19 (s, 3''-CH<sub>3</sub>), 1.18 (d, 5'-CH<sub>3</sub>), 1.12 (d, 2-CH<sub>3</sub>), 1.11 (s, 12-CH<sub>3</sub>), 1.08 (d, 8-CH<sub>3</sub>), 1.04 (d, 4-CH<sub>3</sub>), and 0.79 (t, CH<sub>2</sub>CH<sub>3</sub>).

55 <sup>1</sup>H NMR (CD<sub>3</sub>OD) δ 5.20 (br d, H-13), 4.50 (br d, H-1''), 4.16 (dq, H-5''), 4.02 (d, H-3), 3.70 (m, H-5'), 3.56 (br d, H-5), 3.34 (s, OCH<sub>3</sub>), 3.25 (dd, H-2'), 3.03 (d, H-4''), 2.87 (m, H-8), 2.84 (m, H-2), 2.73 (m, H-3'), 2.44 (d, H-2''eq), 2.33 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.97 (m, H-4), 1.88 (m, H-14eq), 1.73 (m, H-4'eq), 1.64 (m, H-7), 1.59 (dd, H-2''ax), 1.47 (m, H-14ax), 1.36 (br s, 6-CH<sub>3</sub>), 1.28 (d, 5''-CH<sub>3</sub>), 1.24 (s, 3''-CH<sub>3</sub>), 1.18 (m, 5'-CH<sub>3</sub>, 2-CH<sub>3</sub>, 8-

CH<sub>3</sub> and 10-CH<sub>3</sub>)), 1.13 (s, 12-CH<sub>3</sub>), 1.08 (d, 4-CH<sub>3</sub>), and 0.86 (t, CH<sub>2</sub>CH<sub>3</sub>).

<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 176.2, 168.2, 102.8, 95.9, 83.6 (br), 79.3 (br), 77.9, 77.3, 75.2, 75.1, 72.7, 71.0, 70.9, 68.8, 65.5, 65.3, 49.4, 40.2, 39.9 (br), 37.8 (br), 35.7 (br), 34.9, 34.1 (br), 28.9, 26.0 (br), 21.4, 21.3, 19.8 (br), 18.4, 16.8, 15.3 (br), 10.7, and 9.2.

5 <sup>13</sup>C NMR (CD<sub>3</sub>OD) δ 177.7, 170.0, 103.9, 97.7, 84.3 (br), 80.7, 79.2, 78.1, 77.0 (br), 76.1, 74.1, 72.8, 71.7 (br), 69.2, 66.7, 65.1, 49.9, 46.2 (br), 41.8 (br), 40.8, 40.5 (br), 36.0, 33.8 (br), 31.9, 26.7 (br), 22.8, 21.8, 21.7 (br), 21.6, 19.1, 17.5, 15.8 (br), 12.2 (br), 11.3, and 10.1.

FAB mass spectrum, m/z 749, 591, 416, 398, 174, 159, 158, and 116.

10

Elemental Analysis.			
Calculated for C <sub>37</sub> H <sub>68</sub> N <sub>2</sub> O <sub>13</sub> :	C, 59.34;	H, 9.15;	N, 3.74.
Found:	C, 59.12;	H, 8.80;	N, 3.82.

15

#### Method 2: 1.0 LiOH in EtOH

(9E)-9-Deoxo-9-hydroxyiminoerythromycin A (255 mg, 0.34 mmol) was added to a solution of lithium hydroxide monohydrate (14.3 mg, 0.34 mmol) in absolute ethanol (2.55 mL). The resulting solution was stirred at room temperature for 25 hours, and then stored in a freezer at -20 °C for 68 hours. After warming to room temperature, the solution was evaporated under reduced pressure to remove the solvent. The residue was stirred with saturated aqueous sodium chloride (5 mL) and ethyl acetate (5 mL) while the pH was adjusted to 9.2 by addition of dilute hydrochloric acid. After shaking, the phases were separated and the aqueous portion extracted with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate extracts were washed with saturated sodium chloride solution (4 mL), dried over magnesium sulfate, filtered and evaporated at reduced pressure to afford a white foam (263 mg). Examination of this material by <sup>1</sup>H NMR spectroscopy revealed a 31:69 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A.

30

#### Method 3: 2.0 LiOH in EtOH

(9E)-9-Deoxo-9-hydroxyiminoerythromycin A (291 mg, 0.333 mmol) was added to a solution of lithium hydroxide monohydrate (32.6 mg, 0.776 mmol) in absolute ethanol (2.9 mL). The resulting solution was stirred at room temperature under a nitrogen atmosphere for 22.5 hours. The solvent was evaporated at reduced pressure and the residue stirred with ethyl acetate (5 mL) and saturated aqueous sodium chloride (5 mL) while adjusting the pH to 9 by addition of 2N hydrochloric acid. The mixture was shaken, the phases separated, and the aqueous portion extracted with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate extracts were washed with saturated sodium chloride solution (4 mL), dried with magnesium sulfate, filtered and evaporated under vacuum to a white foam (299 mg). This material was shown by <sup>1</sup>H NMR to be a 21:79 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A.

35

#### Method 4: 3.0 LiOH in EtOH

45 (9E)-9-Deoxo-9-hydroxyiminoerythromycin A (239 mg, 0.319 mmol) was added to a mixture of lithium hydroxide monohydrate (40.2 mg, 0.957 mmol) in absolute ethanol (2.4 mL), and the resulting solution was stirred at room temperature under a nitrogen atmosphere for 21.7 hours. Workup as described in method 3 afforded a white foam (236 mg) shown by <sup>1</sup>H NMR to consist of a 19:81 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A.

50

#### Method 5: 2.0 NaOEt in EtOH

55 Freshly cut sodium metal (48 mg, 2.087 mmol) was dissolved in absolute ethanol (7.8 mL) under a nitrogen atmosphere. (9E)-9-Deoxo-9-hydroxyiminoerythromycin A (782 mg, 1.043 mmol) was added and the resulting solution was stirred at room temperature. A crystalline precipitate, identified as the starting oxime by thin layer chromatography, appeared after a few hours. After stirring overnight, the mixture was once again a clear solution. After 54 hours, approximately half (3.9 mL) of the reaction mixture was removed and evaporated under reduced pressure. The gummy residue was stirred with ethyl acetate (5 mL) and

saturated aqueous sodium chloride (5 mL) while the pH was adjusted to 9.2 by addition of dilute hydrochloric acid (2N and 0.2N solutions). The mixture was shaken, the layers separated, and the aqueous portion extracted with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate solution was washed with brine (5 mL), dried with magnesium sulfate, filtered and evaporated under reduced pressure to a white foam (361 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to consist of a 22:78 mixture of the (9E) and (9Z) isomers of 9-deoxo-9-hydroxyiminoerythromycin A.

Method 6: 2.0 NaOH in EtOH

10 The remaining half of the reaction mixture from method 5 was treated with water (0.0188 mL, 1.04 mmol) to give a solution effectively consisting of sodium hydroxide and oxime in ethanol. The solution was stirred at room temperature for 23 hours, then worked up as described in method 5 to give a white foam (402 mg). This material was shown by <sup>1</sup>H NMR to consist of a 24:76 mixture of the (9E) and (9Z) isomers of 9-deoxo-9-hydroxyiminoerythromycin A.

15 Method 7: 2.0 LiOH in MeOH

20 A solution of lithium hydroxide monohydrate (37 mg, 0.88 mmol), (9E)-9-deoxo-9-hydroxyiminoerythromycin A (330 mg, 0.44 mmol), and methanol (3.3 mL) was stirred at room temperature for 65.5 hours. The 25 solution was then stored at -20 °C for 13 days before warming to room temperature and evaporating the solvent at reduced pressure. The residue was stirred with ethyl acetate (5 mL) and brine (5 mL) while adjusting the pH to 9.2 by successive addition of dilute hydrochloric acid. The mixture was shaken, the layers separated and the aqueous portion extracted with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate solution was washed with brine (5 mL), dried with magnesium sulfate, and evaporated under vacuum to provide a white foam (324 mg). NMR analysis of this material indicated a 45:55 ratio of (9E) to (9Z) 9-deoxo-9-hydroxyiminoerythromycin A products.

Method 8: 2.0 NaOMe in MeOH

30 A solution of (9E)-9-deoxo-9-hydroxyiminoerythromycin A (375 mg, 0.5 mmol) in anhydrous methanol (3.5 mL) was cooled in an ice bath and stirred under a nitrogen atmosphere while methanolic sodium methoxide (0.23 mL of a 25 wt % solution; 1.01 mmol) was added by syringe. The cooling bath was removed and the solution was stirred at room temperature under a nitrogen atmosphere for 66 hours. The 35 solution was then stored at -20 °C for 13.3 days before being processed to a white foam (329 mg) as described in method 7. The product consisted of a 35:65 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A as determined by <sup>1</sup>H NMR spectroscopy.

Method 9: 10.0 NaOMe in MeOH

40 A solution of (9E)-9-deoxo-9-hydroxyiminoerythromycin A (100 mg, 0.134 mmol) in anhydrous methanol (4.70 mL) was treated with sodium methoxide (0.305 mL of a 25 wt % solution in methanol, 1.335 mmol) and stirred at room temperature for 74.5 hours. The solvent was evaporated under reduced pressure and the residue stirred with ethyl acetate (5 mL) and brine (5 mL) while adjusting the pH of the aqueous layer to 9.4 with 2N hydrochloric acid. The mixture was shaken, the layers separated and the aqueous portion 45 extracted with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate solution was washed with brine (5 mL), dried with magnesium sulfate, filtered and evaporated at reduced pressure to afford a white foam (102 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to consist of a 26:74 mixture of the (9E) and (9Z) isomers of 9-deoxo-9-hydroxyiminoerythromycin A.

50 Method 10: 2.0 LiOH in iPrOH

(9E)-9-Deoxo-9-hydroxyiminoerythromycin A (279 mg, 0.361 mmol) was added to a partial solution of lithium hydroxide monohydrate (30.3 mg, 0.721 mmol) in isopropanol (2.7 mL), and the mixture was stirred at room temperature in a capped flask. A fine white precipitate formed in a few minutes and, after stirring 55 overnight, the mixture was a hazy suspension. After 21 hours, the mixture was transferred to a freezer at -20 °C and stored there for 15 days. After warming to room temperature, the solvent was evaporated under vacuum and the residue stirred with ethyl acetate (5 mL) and brine (5 mL) while adjusting the pH to 9.2 with dilute hydrochloric acid. The mixture was shaken, the layers separated, and the aqueous phase extracted

with more ethyl acetate (2 x 2.5 mL). The combined ethyl acetate solution was washed with brine (4 mL), dried with magnesium sulfate, filtered and evaporated under vacuum to afford a white foam (249 mg). The product consisted of a 26:74 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A as determined by <sup>1</sup>H NMR spectroscopy.

### Method 11: 1.0 LiOH in MeCN

10 A mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A (500 mg, 0.668 mmol), lithium hydroxide monohydrate (28 mg, 0.668 mmol), and absolute ethanol (5 mL) was stirred at room temperature for 10 minutes to give a solution. The solution was evaporated under reduced pressure to afford a residue that was twice diluted with ethanol (10 mL) and evaporated at reduced pressure and then suspended in anhydrous acetonitrile (5 mL) and evaporated at reduced pressure. The solid residue was suspended in anhydrous acetonitrile (5 mL) and the mixture was stirred at room temperature for 18 days. The solvent was evaporated under reduced pressure and the residue was stirred with ethyl acetate (5 mL) and saturated aqueous sodium chloride solution (5 mL) while adjusting the pH of the aqueous phase to 9.5 by addition of dilute hydrochloric acid. The mixture was shaken, the layers separated, and the aqueous portion was extracted with additional ethyl acetate (2 x 2.5 mL). The combined ethyl acetate solution was washed with brine (5 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure to afford a foam (442 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to consist of a 44:56 mixture of the (9E) and (9Z) isomers of 9-deoxo-9-hydroxyiminoerythromycin A.

### Method 12: 1.0 M LiOH in DMF

25 A mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A (500 mg, 0.668 mmol), lithium hydroxide monohydrate (28 mg), and dimethylformamide (5 mL) was stirred at room temperature in a capped flask. After a few hours, the initial suspension gave way to a solution. After stirring for 18 days and 18 hours, the solution was evaporated under reduced pressure and the residue was processed as described in method 11 to afford a foam (402 mg). Analysis of this material by  $^1\text{H}$  NMR spectroscopy indicated a 62:38 mixture of the (9E) and (9Z) isomers of 9-deoxo-9-hydroxyiminoerythromycin A.

### Method 13: 1.2 LiN(SiMe<sub>3</sub>)<sub>2</sub> in MeCN

35 A suspension of (9E)-9-deoxo-9-hydroxyiminoerythromycin (500 mg, 0.668 mmol) in anhydrous acetonitrile (4 mL) was treated with lithium hexamethyldisalazide (0.80 mL of a 1M solution in hexane, 0.80 mmol). The resulting suspension rapidly gave way to a solution which reformed a suspension after stirring several days at room temperature. After 18 days and 19 hours, the reaction mixture was worked up as described in method 11 to afford a foam (423 mg). This material was shown by  $^1\text{H}$  NMR spectroscopy to be a 50:50 mixture of (9E)-9-deoxo-9-hydroxyiminoerythromycin A and (9Z)-9-deoxo-9-hydroxyiminoerythromycin A.

**40 EXAMPLE 3**

## Crystallization of (9Z)-9-Deoxo-9-hydroxyiminoerythromycin A

45 A 3:1 mixture (30.0 g) of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A and (9E)-9-deoxo-9-hydroxyiminoerythromycin A was added over 2 minutes to well stirred ethyl acetate (60 mL). After obtaining a solution, methylene chloride (120 mL) was rapidly added and the resulting suspension was stirred in an ice bath for one hour. The precipitate was filtered off, washed with methylene chloride (60 mL), and dried under a stream of nitrogen to afford an 86:14 mixture (26.5 g) of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A and (9E)-9-deoxo-9-hydroxyiminoerythromycin A.

50 A solution of the above solid in ethyl acetate (60 mL) was diluted with methylene chloride (120 mL). The resulting suspension was cooled in an ice bath for one hour and then filtered. The collected solid was rinsed with methylene chloride (60 mL) and dried under a stream of nitrogen to afford a 95:5 mixture (23.4 g) of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A and (9E)-9-deoxo-9-hydroxyiminoerythromycin A.

## EXAMPLE 4

5

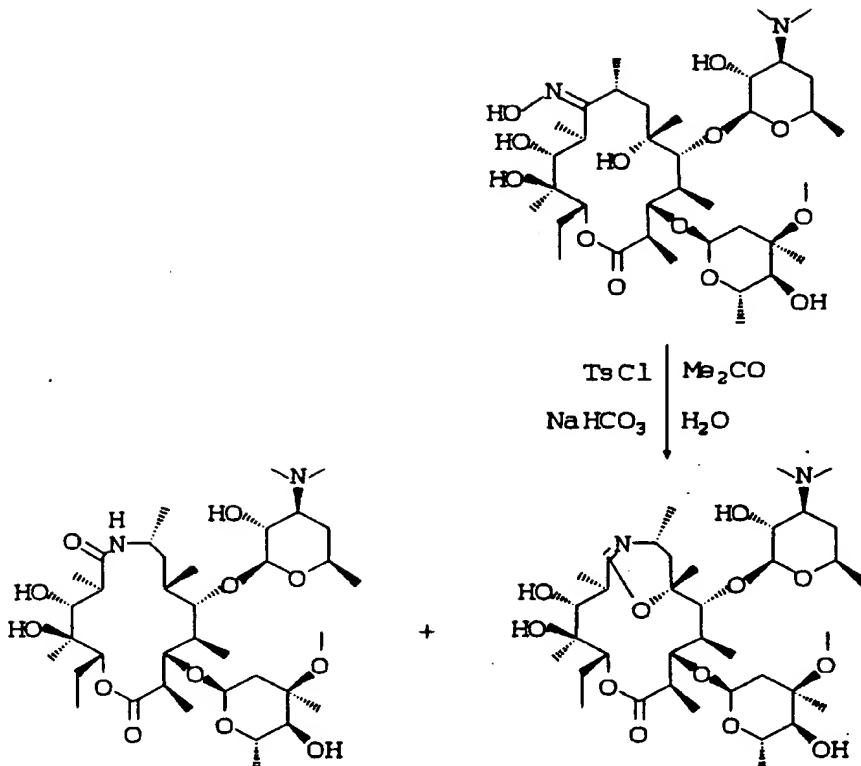
10

15

20

25

30



Synthesis of 8a-Aza-8a-homoerythromycin A and 9-Deoxy-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A by the Beckmann Rearrangement of 9Z-9-Deoxo-9-hydroxyiminoerythromycin A

35

Method 1:

(9Z)-9-Deoxo-9-hydroxyiminoerythromycin A (200 mg, 9.27 mMol) was dissolved in acetone (2 mL) and the resulting solution was cooled in an ice-bath and stirred under a nitrogen atmosphere. A solution of sodium bicarbonate (84 mg, 1.0 mMol) in water (2 mL) was added followed by the dropwise addition of an acetone solution (2 mL) of p-toluenesulfonyl chloride (100 mg, 0.53 mMol) over 5 minutes.

After stirring for 1.5 hours at 0-5 °C, the mixture was diluted with dichloromethane (10 mL) and water (5 mL), and the pH was adjusted from 10 to 5.5 with 2N HCl. The dichloromethane layer was discarded and the aqueous layer was washed with additional dichloromethane (2 x 10 mL) which was also discarded. Dichloromethane (10 mL) was added to the aqueous layer and the pH was adjusted to 8.5 with 2.5 N NaOH. The dichloromethane layer was removed and the aqueous layer was extracted with more dichloromethane (2 x 20 mL). The combined dichloromethane extracts were dried over anhydrous magnesium sulfate, filtered and evaporated under vacuum to give a mixture of the title compounds as a foam (150 mg).

The above mixture was purified by preparative layer chromatography (two 0.1 mm x 20 x 20 cm Analtech silica gel GF plates, developing and eluting with 60:10:1 dichloromethane-methanol concentrated ammonium hydroxide) to afford 8a-aza-8a-homoerythromycin A (95 mg.) and 9-deoxy-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (33 mg).

Method 2:

55

A solution of p-toluenesulfonyl chloride (1.00 g, 5.2 mmol) in acetone (20 mL) was added to a solution of sodium bicarbonate (0.90 g, 10.7 mmol) in water (20 mL). The resulting suspension was cooled in a -10 °C bath and stirred while a solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (2.00 g, 2.7 mmol) in

acetone (20 mL) was added slowly over 75 minutes. The mixture was stirred at -10 °C for 5 hours, then warmed to 0 °C over 10 minutes and stirred at 0-5 °C for 30 minutes. The mixture was evaporated under vacuum to remove the acetone. The aqueous residue was diluted with water (40 mL) and dichloromethane (60 mL) and stirred while the pH was adjusted to 5.5 with dilute hydrochloric acid. The aqueous layer was separated, washed with dichloromethane (60 mL), layered with dichloromethane (60 mL), and stirred while the pH was brought to 9 with dilute aqueous sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (2 x 50 mL). The combined pH 9 extracts were dried over magnesium sulfate, filtered and evaporated under reduced pressure to afford a gum (1.97 g) which was shown by <sup>1</sup>H NMR spectroscopy to be a 1:1 mixture of 8a-aza-8a-homoerythromycin A and 9-deoxy-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

The crude product mixture was dissolved in 120:10:1 dichloromethane-methanol-conc. aqueous ammonium hydroxide (5 mL) and loaded onto a column of silica gel (4 x 16 cm). The column was eluted with 120:10:1 dichloromethane-methanol-ammonium hydroxide. After a 150 mL forerun, 15 mL fractions were collected. Fractions 9-13 were combined and evaporated under vacuum to afford 9-deoxy-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (ca. 500 mg) and fractions 22-26 were combined and evaporated to afford 8a-aza-8a-homoerythromycin A (ca. 500 mg). The later product was crystallized from ether to give the amide (ca. 130 mg) as a white solid.

Physical data for 9-deoxy-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A:

IR (CHCl<sub>3</sub>) 3550, 3440 (br), 2970, 2940, 2880, 1725, 1665, 1455, 1375, 1345, 1325, 1240, 1170, 1105, 1080, 1050, 1015, 995, and 955 cm<sup>-1</sup>.  
<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.02 (d, H-1"), 4.90 (dd, H-13), 4.48 (d, H-1'), 4.09 (dq, H-5"), 4.02 (t, H-3), 3.81 (d, H-5), 3.53 (m, H-5'), 3.49 (d, H-11), 3.43 (m, H-8), 3.35 (s, OCH<sub>3</sub>), 3.20 (dd, H-2'), 3.07 (t, H-4"), 2.75 (dq, H-2), 2.68 dq, H-10), 2.52 (ddd, H-3'), 2.43 (d, H-2"eq), 2.28 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.98 ddq, H-4), 1.91 (m H-14a), 1.90 (dd, H-7a), 1.68 (ddd, H-4"eq), 1.62 (dd, H-2"ax), 1.46 (m, H-14b), 1.39 (s, 6-CH<sub>3</sub>), 1.32 (d, 5"-CH<sub>3</sub>), 1.27 (s 3"-CH<sub>3</sub>), 1.24 (m, H-7b), 1.22 (d, 5'-CH<sub>3</sub>), 1.21 (m H-4'ax), 1.16 (d, 10-CH<sub>3</sub>), 1.15 d, 8-CH<sub>3</sub>), 1.15 (s, 12-CH<sub>3</sub>), 1.14 (d, 2-CH<sub>3</sub>), 1.08 (d, 4-CH<sub>3</sub>), and 0.87 (t, CH<sub>2</sub>CH<sub>3</sub>).  
<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 177.6, 160.6, 102.4, 94.6, 80.1, 78.9, 77.9, 77.4, 76.5, 75.7, 73.0, 70.6, 70.0, 68.8, 65.8, 65.6, 49.4, 44.9, 44.0, 42.3, 42.1, 40.3, 34.5, 32.0, 28.5, 23.8, 22.4, 21.5, 21.3, 21.0, 18.2, 17.0, 16.4, 12.5, 10.8, and 8.4.  
FAB mass spectrum, m/z 731, 713, 602, 573, 555, 398, 159, 158, and 116.

Physical data for 8a-aza-8a-homoerythromycin A:

MP 170-176 °C.  
IR (CHCl<sub>3</sub>) 3500 (br), 3430, 3320, 2970, 2935, 2880, 17730, 1630, 1560, 1525, 1455, 1375, 1325, 1280, 1170, 1160, 1105, 1085, 1045, 1010 and 995 cm<sup>-1</sup>.  
<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.89 (br d, NH), 5.07 (d, H-1"), 4.92 (dd, H-13), 4.43 (d, H-1'), 4.35 (d, H-3), 4.21 (m, H-8), 4.01 (dq, H-5"), 3.58 (d, H-5), 3.50 (m, H-5'), 3.50 (s, H-11), 3.32 (s, OCH<sub>3</sub>), 3.21 (dd, H-2'), 3.03 (t, H-4"), 2.62 (dq, H-2), 2.54 (m, H-3'), 2.35 (m, H-10), 2.35 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.31 (d, H-2"eq), 1.90 (m, H-4), 1.89 (m, H-14a), 1.75 (br d, H-4"eq), 1.57 (dd, H-2"ax), 1.51 (m, H-7a and H-7b), 1.44 (m, H-14b), 1.43 (s, 6-CH<sub>3</sub>), 1.30 (d, 5"-CH<sub>3</sub>), 1.24 (s, 3"-CH<sub>3</sub>), 1.23 (m, H-4'ax), 1.23 (d, 5'-CH<sub>3</sub>), 1.20 (d, 8-CH<sub>3</sub>), 1.19 (d, 10-CH<sub>3</sub>), 1.18 (d, 2-CH<sub>3</sub>), 1.09 (s, 12-CH<sub>3</sub>), 1.05 (d, 4-CH<sub>3</sub>), and 0.89 (t, CH<sub>2</sub>CH<sub>3</sub>).  
<sup>13</sup>C NMR (CDCl<sub>3</sub>) 177.6, 176.6, 102.7, 94.2, 83.0 77.9, 77.0, 76.6, 74.6, 73.7, 72.9, 70.0, 69.8, 68.8, 65.8, 65.2, 49.2, 45.8, 43.2, 42.4, 41.0, 40.4, 40.1, 34.5, 28.3, 27.6, 23.1, 21.7, 21.5, 21.2, 18.0, 16.1, 14.6, 11.2, 10.0, and 9.1.  
FAB mass spectrum, m/z 749, 731, 591, 589, 573, 416, 174, 159, 158, and 117.

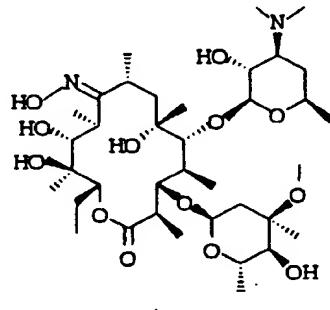
Elemental Analysis.			
Calculated for C <sub>37</sub> H <sub>68</sub> N <sub>2</sub> O <sub>13</sub> :	C, 59.31;	H, 9.15;	N, 3.74.
Found:	C, 59.24;	H, 9.15;	N, 3.44

Loss on drying at 120 °C, 3.11%.

## EXAMPLE 5

Synthesis of 9-Deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A by Beckmann Rearrangement of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A

10

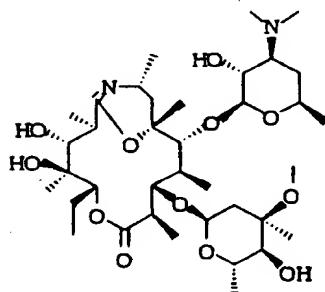


15

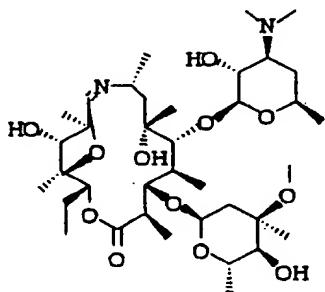
$\xrightarrow[\text{C}_2\text{H}_5\text{N}]{\text{RSO}_2\text{Cl}}$

20

25



30

35 Method 1:

A solution of p-toluenesulfonyl chloride (15.0 g, 0.079 mol) in diethyl ether (50 mL) was added dropwise over 8 minutes to an ice-cold, stirring solution of 9(Z)-9-deoxo-9-hydroxyiminoerythromycin A (23.2 g, 0.031 mol) in pyridine (180 mL). The resulting solution was stirred at 0-5 °C for 2.5 hours, then diluted with dichloromethane (400 mL) and water (500 mL) and basified to pH 9.5 by addition of 5N sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (200 mL, 100 mL). The combined dichloromethane extracts were dried over magnesium sulfate, filtered, and evaporated under vacuum to afford an oil. Residual pyridine was removed by twice taking the product up in toluene (100 mL) and evaporating the solvent under vacuum. The resulting foam (21.4 g) was shown by 1H NMR spectroscopy to be a 26:74 mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 2:

50 A solution of p-toluenesulfonyl chloride (160 mg, 0.84 mmol) in diethyl ether (0.5 mL) was added rapidly to an ice-cold solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL). The resulting solution was stirred at 0-5 °C for 1.5 hours, then diluted with dichloromethane (4 mL) and water (4 mL) and basified to pH 9.5 by addition of 5N sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (2 x 4 mL). The combined dichloromethane extracts 55 were dried over magnesium sulfate, filtered, evaporated under vacuum and stripped with hexane (4 x 15 mL) to afford a yellow solid (260 mg.) This material was shown by 1H NMR spectroscopy to be a 25:75 mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 3:

5 A solution of p-toluenesulfonyl chloride (160 mg, 0.84 mmol) in acetonitrile (0.5 mL) was added rapidly to an ice-cold solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL). The resulting solution was stirred at 0-5 °C for 80 minutes, then diluted with dichloromethane (4 mL) and water (5 mL) and basified to pH 9.5 by addition of 5N sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (2 x 4 mL). The combined dichloromethane extracts were dried over magnesium sulfate, filtered, and evaporated under vacuum to a foam which was stripped with toluene (2 x 10 mL) and hexanes (10 mL) to afford a solid (230 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to be a 33:67 mixture of 9-deoxo-6-deoxy-6,9-epoxy-and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 4:

15 A solution of p-toluenesulfonyl chloride (160 mg, 0.84 mmol) in toluene (0.5 mL) was added rapidly to an ice-cold solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL). The resulting solution was stirred at 0-5 °C for 90 minutes, then diluted with dichloromethane (4 mL) and water (4 mL) and basified to pH 9.5 by addition of 1N sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (3 x 4 mL). The combined dichloromethane extracts were dried over magnesium sulfate, filtered, and evaporated under vacuum to a solid (250 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to be a 27:73 mixture of 9-deoxo-6-deoxy-6,9-epoxy-and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 5:

25 Benzenesulfonyl chloride (0.107 mL, 0.84 mmol) was added by syringe to an ice-cold solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL). The resulting solution was stirred at 0-5 °C for 75 minutes, then processed as described above to afford a yellow solid (240 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to be a 31:69 mixture of 9-deoxo-6-deoxy-6,9-epoxy-and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 6:

35 Methanesulfonyl chloride (0.065 mL, 0.84 mmol) was added by syringe to an ice-cold solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL). The resulting solution was stirred at 0-5 °C for 2 hours, then processed as described above to afford an off-white solid (246 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to be a 25:70:5 mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A, 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A, and 9-deoxy-12-deoxy-9,12-epoxy-4"-0-methanesulfonyl-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Method 7:

45 A solution of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (250 mg, 0.33 mmol) in pyridine (2.0 mL) was cooled in a -20 °C bath and treated with methanesulfonyl chloride (0.071 mL, 0.92 mmol). The resulting hazy solution was stirred at -10 to -20 °C for 90 minutes, then processed as described above to afford a yellow solid (254 mg). This material was shown by <sup>1</sup>H NMR spectroscopy to be a 88:12 mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

50

Method 8:

55 A mixture of (9Z)-9-deoxo-9-hydroxyiminoerythromycin A (0.50 g, 0.67 mmol), p-toluenesulfonyl chloride (318 mg, 1.67 mmol) and pyridine (0.162 mL, 2.0 mmol) in dichloromethane (5.0 mL) was stirred at room temperature for 1.5 hours. The mixture was diluted with water and stirred rapidly while adjusting the pH to 11 with 5N sodium hydroxide. The organic phase was separated, dried with magnesium sulfate, filtered and evaporated under reduced pressure to afford a yellow solid (570 mg.) Analysis of the crude product by <sup>1</sup>H NMR spectroscopy revealed a 80:20 mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-

8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9, 12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Example 6

Purification of 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A by Column Chromatography

Although unnecessary for the following reduction step, it is possible to separate the isomeric 6,9-epoxy-10 and 9,12-epoxy products by silica gel or alumina column chromatography. The following procedure illustrates the purification process for 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

The crude products from methods 3 and 4 above were combined, dissolved in 94:5:1 dichloromethane-methanol-triethylamine, and loaded onto a column of silica gel (230-400 mesh, 2.5 x 24.5 cm, wet packed under 94:5:1 dichloromethane-methanol-triethylamine). The column was eluted with 94:5:1 dichloromethane-methanol-triethylamine, collecting 6 mL fractions. Fractions 15-18 were combined, evaporated under reduced pressure, and the residue twice stripped with toluene to provide 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (190 mg) as a foam. The product is a mixture of major and minor forms, isomeric about the 8a,9-imino double bond, as ascertained by <sup>1</sup>H and <sup>13</sup>C NMR spectroscopy.

IR (CHCl<sub>3</sub>) 3550, 3390 (br), 2975, 2940, 2880, 1735, 1690, 1455, 1375, 1240, 1165, 1085, 1045, 1010, and 970 cm<sup>-1</sup>

FAB mass spectrum, m/z 731, 713, 602, 573, 556, and 158

Example 7

Chromatographic Separation of 9-Deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and Crystallization of 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A

A sample (4.0 g) of the crude product mixture obtained as described in method 1 of Example 5 was dissolved in 60:10:1 dichloromethane-methanol-conc. aqueous ammonium hydroxide (6 mL) and the solution was loaded onto a column of EM silica gel 60 (4.5 x 18 cm, 230-400 mesh, wet packed under 60:10:1 dichloromethane-methanol-conc. ammonium hydroxide). The column was eluted with 60:10:1 dichloromethane-methanol-conc. aqueous ammonium hydroxide. The fractions collected from 150 mL to 165 mL of eluant were evaporated under vacuum to afford 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (0.34 g) as a foam. The fractions collected from 185 mL to 285 mL of eluant were combined and evaporated under reduced pressure to afford a mixture of the two isomeric forms of 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (1.36 g) as a foam.

A solution of the mixture of 9,12-epoxy isomers in nitromethane (2 mL) deposited a large, crystalline mass on standing at room temperature for several days. The mixture was diluted with nitromethane (10 mL) and filtered to remove the solid portion, which was washed with nitromethane (2 mL) and dried under high vacuum. The white solid thus obtained (0.9 g) was shown by <sup>1</sup>H NMR spectroscopy to be the major 9,12-epoxy isomer which is initially formed in the Beckmann rearrangement reaction. While stable in the solid state, solutions of the crystalline isomer in chloroform-d isomerize at room temperature in several hours to a 1:1 mixture of the two imino double bond isomers of 9-deoxo-12-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A.

Physical data for 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A:

50 Isomer A (crystalline isomer)

MP 124-130 °C (slowly softens).

IR (CHCl<sub>3</sub>) 3550, 3380 (br), 2970, 2935, 2875, 1735, 1695, 1560, 1460, 1375, 1250, 1165, 1115, 1058, 1045, 1015, and 975 cm<sup>-1</sup>.

<sup>1</sup>H NMR (CDCl<sub>3</sub>)  $\delta$  5.17 (dd, H-13), 4.73 (d, H-1''), 4.47 (d, H-1'), 4.15 (dq, H-5''), 4.09 (dd, H-3), 3.99 (br s, H-5), 3.81 (t, H-11), 3.68 (m, H-8), 3.65 (m, H-5'), 3.40 (ddd, H-2'), 3.23 (s, OCH<sub>3</sub>), 2.96 (t, H-4''), 2.70 (0, H-10), 2.68 (m, H-3'), 2.57 (br d, 11-OH), 2.45 (p, H-2), 2.31 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.28 (d, H-2''eq), 2.20 (d, 4''-OH), 2.07 (ddq, H-14a), 1.90 (br d, H-7a), 1.75 (dd, H-7b), 1.74 (m, H-4), 1.70 (m, H-4''eq), 1.69 (m, H-14b), 1.46

(dd, H-2"az), 1.40 (s, 6-CH<sub>3</sub>), 1.29 (m, H-4'ax), 1.27 (d, 10-CH<sub>3</sub>), 1.27 (d, 5"-CH<sub>3</sub>), 1.25 (d, 2-CH<sub>3</sub>), 1.24 (d, 5'-CH<sub>3</sub>), 1.21 (s, 3"-CH<sub>3</sub>), 1.18 (s, 12-CH<sub>3</sub>), 1.07 (d, 8-CH<sub>3</sub>), 1.01 (d, 4-CH<sub>3</sub>), and 0.86 (t, CH<sub>2</sub>CH<sub>3</sub>).

<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 174.2, 161.3, 106.7, 98.3, 85.4, 84.2, 80.5, 79.8, 77.4, 75.0, 72.3, 70.3, 69.4, 66.3, 63.8, 49.4, 49.2, 49.0, 47.1, 45.4, 43.2, 40.4, 35.0, 29.3, 27.5, 24.6, 24.4, 23.3, 21.4, 21.0, 17.6, 17.2, 16.9, 11.3, and 11.2.

Elemental Analysis. Calculated for C<sub>37</sub>H<sub>66</sub>N<sub>2</sub>O<sub>12</sub>: C, 60.80; H, 9.10; N, 3.83. Found: C, 60.71; H, 9.38; N, 3.78. Loss on drying at 120 °C, 2.82%.

Isomer B

<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.20 (dd, H-13), 4.74 (d, H-1"), 4.48 (d, H-1'), 4.17 (t, H-3), 4.15 (m H-5"), 4.11 (dd, H-11), 3.97 (m, H-8), 3.71 (d, H-5), 3.62 (m, H-5'), 3.30 (br dd, H-2'), 3.23 (s, OCH<sub>3</sub>), 2.97 (t, H-4"), 2.88 (d, 11-OH), 2.85 (p, H-10), 2.60 (m, H-3'), 2.46 (p, H-2), 2.28 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.27 (d, H-2"eq), 2.23 (d, 4"-OH), 1.98 (ddq, H-14a), 1.84 (dd, H-7a), 1.77 (m, H-4), 1.76 (m, H-14b), 1.66 (m, H-4"eq), 1.64 (dd, H-7b), 1.49 (dd, H-2"ax), 1.29 (s, 6-CH<sub>3</sub>), 1.27 (d, 5"-CH<sub>3</sub>), 1.19 (d, 10-CH<sub>3</sub>), 1.19 (s, 3"-CH<sub>3</sub>), 1.14 (s, 12-CH<sub>3</sub>), 1.09 (d, 8-CH<sub>3</sub>), 1.09 (d, 4-CH<sub>3</sub>), and 0.94 (t, CH<sub>2</sub>CH<sub>3</sub>).

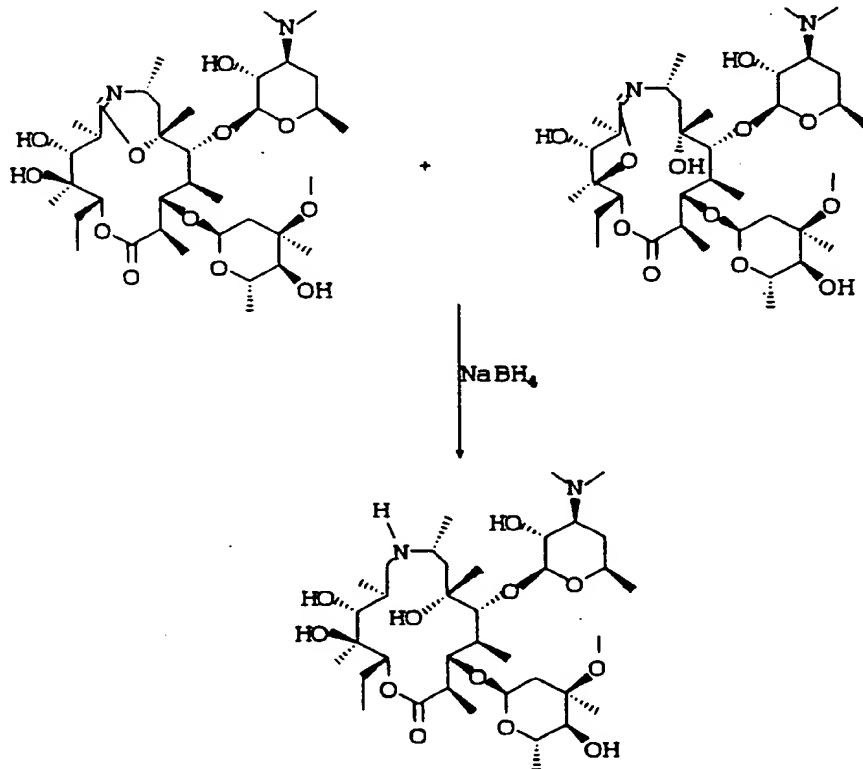
<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 174.4, 160.5, 104.6, 97.0, 86.2, 79.1, 78.6, 77.7, 77.4, 75.1, 70.5, 69.4, 66.0, 64.7, 49.4, 48.2, 47.7, 47.4, 42.3, 40.4, 34.9, 29.1, 25.6, 24.0, 23.6, 22.9, 21.5, 21.0, 15.8, 11.7, 10.7, and 9.6.

Example 8

Synthesis of 9-Deoxo-8a-aza-8a-homoerythromycin A by Sodium Borohydride Reduction of 9-Deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-Deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A

25

30



55

Method 1:

A solution of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a homoerythromycin A (22.6 g of a 27:73 mixture, 0.031 mol) in methanol (50 mL) was cooled in an ice bath and stirred under a nitrogen atmosphere. Solid sodium borohydride (3.6 g, 0.093 mol) was added in portions over 3 hours. The resulting, viscous solution was allowed to slowly warm to room temperature and then stirred at room temperature overnight. The solution was diluted with water (50 mL), acidified to pH 2 with 2N hydrochloric acid, and stirred at room temperature for 10 minutes. The solution was diluted with water (150 mL) and dichloromethane (200 mL) and stirred vigorously while the pH was brought to 6.5 by addition of 5N sodium hydroxide. The dichloromethane layer was discarded and the aqueous phase layered with fresh dichloromethane, stirred rapidly and basified to pH 9.5 with 5N sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (2 x 100 mL). The combined pH 9.5 dichloromethane extracts were dried over magnesium sulfate, filtered and evaporated under reduced pressure to a foam (15.4 g).

The crude product was dissolved in 2-propanol (90 mL) and stirred at room temperature to give a crystalline precipitate. This material was collected, washed with cold 2-propanol (20 mL) and dried to afford 9-deoxo-8a-aza-8a-homoerythromycin A (6.0 g) as a white solid.

The mother liquors and washings were evaporated under vacuum to a solid residue. The solid was suspended in water (50 mL), acidified to pH 2, and stirred at room temperature for 30 minutes. The mixture was diluted with water (50 mL) and dichloromethane (100 mL), then stirred vigorously while adjusting the pH to 6.5. The dichloromethane layer was discarded and replaced with fresh dichloromethane (100 mL). The mixture was stirred while the pH was adjusted to 9.5. The layers were separated and the aqueous phase was extracted with more dichloromethane (2 x 100 mL). The combined basic extracts were dried with magnesium sulfate, filtered and evaporated under vacuum to a foam (6.2 g). This material was dissolved in 2-propanol (30 mL) and the solution cooled in ice to give additional crystalline product. The solid was collected and dried to afford additional 9-deoxo-8a-aza-8a-homoerythromycin A (2.7 g).

MP 177-180 °C  
 IR (CHCl<sub>3</sub>) 3540, 3340 (br), 2970, 2930, 2880, 1725, 1450, 1375, 1325, 1125, 1105, 1085, 1065, 1045, 955, and 870 cm<sup>-1</sup>.  
<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.00 (d, H-1"), 4.75 (dd, H-13), 4.48 (br d, H-3), 4.34 (d, H-1'), 4.02 (dq, H-5"), 3.56 (br s, H-11), 3.52 (d, H-5), 3.45 (m, H-5'), 3.31 (s, OCH<sub>3</sub>), 3.16 (dd, H-2'), 3.01 (br d, H-4"), 2.78 (m, H-8), 2.69 (dq, H-2), 2.59 dd, H-9a), 2.42 (br t, H-9b), 2.30 (d, H-2"eq), 2.26 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.91 (m, H-14a), 1.77 (br p, H-4), 1.61 (br d, H-4"eq), 1.55 (dd, H-2"ax), 1.44 (m, H-14b), 1.38 (m, H-7), 1.36 (s, 6-CH<sub>3</sub>), 1.29 (d, 5"-CH<sub>3</sub>), 1.21 (s, 3"-CH<sub>3</sub>), 1.20 (d 5'-CH<sub>3</sub>), 1.18 (d, 2-CH<sub>3</sub>), 1.10 (d, 8-CH<sub>3</sub>), 1.06 (s 12-CH<sub>3</sub>), 1.04 (d, 4-CH<sub>3</sub>), 0.94 (d, 10-CH<sub>3</sub>), and 0.86 (t, CH<sub>2</sub>CH<sub>3</sub>).  
<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 178.6, 103.4, 94.6, 83.6, 78.1, 77.6, 76.6, 74.9, 72.8, 70.7, 68.9, 66.8, 65.7, 65.2, 49.6, 49.4, 45.5, 43.4, 40.3, 35.3, 34.7, 28.7, 27.6, 21.6, 21.3, 20.8, 18.2, 16.3, 15.1, 12.1, 11.3, and 9.5.  
 FAB mass spectrum, m/z 735, 577, 559, 402, 159, 158, and 116.

40	Elemental Analysis.			
	Calculated for C <sub>37</sub> H <sub>70</sub> N <sub>2</sub> O <sub>12</sub> :	C, 60.47;	H, 9.60;	N, 3.81.
	Found:	C, 59.98;	H, 9.46;	N, 3.62.

45 Loss on drying at 120 °C, 0.33%.

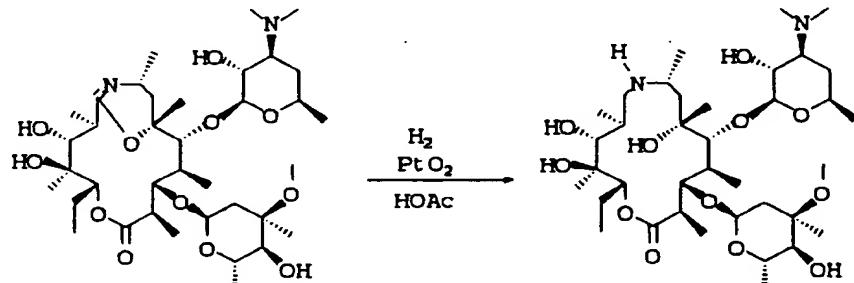
Method 2:

A solution of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A and 9-deoxo-12-deoxy-9,12-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (5.0 g of a 1:3 mixture, 6.84 mmol) in ethylene glycol (25 mL) was cooled in an ice bath and stirred under a slow stream of nitrogen. Sodium borohydride (0.60 g, 15.86 mmol) was added in two nearly equal portions spaced one hour apart. Following the borohydride addition, the reaction mixture was stirred at 0-5 °C for 1.5 hours, then warmed to room temperature and stirred at room temperature overnight. The reaction mixture was diluted with water (50 mL) and dichloromethane (25 mL), stirred vigorously, and phases separated. The aqueous portion was extracted with more dichloromethane (4 x 25 mL). The combined organic extracts were washed with brine (50 mL), dried over magnesium sulfate, filtered and evaporated under reduced pressure to foam (4.0 g).

The crude product was dissolved in 2-propanol (20 mL) and the solution stirred at room temperature to give a crystalline precipitate. The product was collected and dried under a stream of nitrogen to afford 9-deoxo-8a-aza-8a-homoerythromycin A (2.2 g) as a white solid.

5 EXAMPLE 9
Synthesis of 9-Deoxo-8a-aza-8a-homoerythromycin A by Catalytic Hydrogenation of 9-Deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A

10



15

20

A mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (100 mg), acetic acid (4 mL) and platinum oxide (120 mg) was hydrogenated overnight at 2000 psi. The mixture was filtered through celite and the filtrate evaporated under vacuum to a residue that was partitioned between dichloromethane (12 mL) and saturated aqueous sodium bicarbonate (5 mL). The dichloromethane layer was removed and the aqueous layer was extracted with more dichloromethane (2 x 5 mL). The combined dichloromethane extracts were dried over anhydrous magnesium sulfate, filtered and evaporated under vacuum to a gum (60 mg).

The oil was purified by preparative thin layer chromatography (Analtech 0.1 mm x 20 x 20 cm basic alumina plate, developing and eluting with 5% methanol in dichloromethane) to give the title compound as a white foam (42 mg).

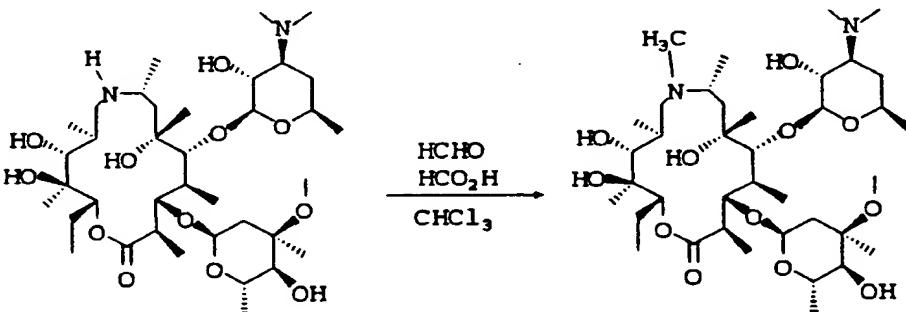
In a manner similar to that described above, a mixture of 9-deoxo-6-deoxy-6,9-epoxy-8a,9-didehydro-8a-aza-8a-homoerythromycin A (400 mg) and platinum oxide (400 mg) in acetic acid (8 mL) was hydrogenated at 2000 psi overnight. Workup provided a foam (372 mg) that was dissolved in 1% methanolic dichloromethane (2 mL) and loaded onto a column of basic alumina (1.75 x 26 cm, wet packed using 1% methanol in dichloromethane). The column was eluted with 1% methanol in dichloromethane, collecting 6 mL fractions. Fractions 27-42 were combined and evaporated under reduced pressure to afford 9-deoxo-8a-aza-8a-homoerythromycin A (178 mg) as a foam.

40

EXAMPLE 10
Synthesis of 9-Deoxo-8a-aza-8a-methyl-8a-homoerythromycin A by Methylation of 9-Deoxo-8a-aza-8a-homoerythromycin A

45

50



A solution of 9-deoxo-8a-aza-8a-homoerythromycin A (7.30 g, 9.9 mmol) in chloroform (45 mL) was treated with 37% aqueous formaldehyde (0.81 mL, 10.8 mmol) and 98% formic acid (1.08 mL, 28.0 mmol). The resulting mixture was heated at reflux for 25.5 hrs., then cooled to room temperature, diluted with dichloromethane (150 mL) and water (120 mL), and stirred vigorously for a few minutes. The dichloromethane layer was discarded and fresh dichloromethane (100 mL) was added. The mixture was stirred rapidly while the pH was adjusted to 9.5 by addition of 5N sodium hydroxide. The dichloromethane layer was removed and the aqueous portion was re-extracted with more dichloromethane (50 mL, 25 mL). The combined dichloromethane extracts were dried over anhydrous magnesium sulfate, filtered and evaporated under vacuum to a foam (7.27 g).

10 A solution of the foam in warm ethanol (24 mL) was diluted with water (12 mL) and stirred at room temperature for 5 minutes to give a precipitate. The mixture was diluted with more water (12 mL), stirred with ice-bath cooling, and then left in a refrigerator overnight. The mixture was filtered and the collected solid was rinsed with cold 3:1 water-ethanol (12 mL), dried under a stream of nitrogen, and finally dried under vacuum to afford the title compound (6.20 g) as a white solid.

15 MP 187-188 °C.

IR (CHCl<sub>3</sub>) 3540, 3330 (br), 2970, 2940, 2880, 2830, 1725, 1455, 1375, 1350, 1325, 1275, 1160, 1125, 1105, 1085, 1065, 1045, 995, 975, and 955 cm<sup>-1</sup>.

<sup>1</sup>H NMR (CDCl<sub>3</sub>, 55 °C) δ 5.10 (d, H-1"), 4.86 (dd, H-13), 4.51 (t, H-3), 4.38 (d, H-1'), 4.04 (dq, H-5"), 3.53 (br s, H-11), 3.52 (d, H-5), 3.51 (m, H-5'), 3.32 (s, OCH<sub>3</sub>), 3.23 (dd, H-2'), 3.01 (dd, H-4"), 2.99 (m, H-8), 2.81 (dq, H-2), 2.52 (m, H-9a), 2.40 (m, H-3'), 2.34 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.30 (m, H-9b), 2.30 (d, H-2"eq), 2.04 (s, NCH<sub>3</sub>), 1.99 (m, H-10), 1.92 (m, H-14a), 1.88 (m, H-7a), 1.85 (m, H-4), 1.72 (br d, H-4'eq), 1.55 (dd, H-2"ax), 1.48 (m, H-14b), 1.37 (s, 6-CH<sub>3</sub>), 1.30 (d, 5" -CH<sub>3</sub>), 1.24 (d, 5'-CH<sub>3</sub>), 1.23 (m, H-4'ax), 1.23 (s, 3"-CH<sub>3</sub>), 1.19 (d, 2-CH<sub>3</sub>), 1.12 (m, H-7b), 1.10 (d, 4-CH<sub>3</sub>), 1.10 (s, 12-CH<sub>3</sub>), 0.96 (d, 10-CH<sub>3</sub>), 0.94 (d, 8-CH<sub>3</sub>), and 0.92 (t, CH<sub>2</sub>CH<sub>3</sub>).

25 <sup>13</sup>C NMR (CDCl<sub>3</sub>, 55 °C) δ 178.3, 103.6, 94.7, 85.5, 78.4, 77.2, 76.7, 75.9, 74.9, 73.1, 71.0, 69.1, 67.1, 65.8, 65.4, 60.0, 56.7, 49.4, 45.8, 43.5, 40.4, 37.1, 35.1, 30.9, 29.3, 27.8, 22.1, 21.7, 21.3, 18.3, 16.4, 14.3, 12.7, 12.0, 11.4, and 11.3.

FAB mass spectrum, m/z 749, 591, 573, 158, and 116.

30

Elemental Analysis.			
Calculated for C <sub>38</sub> H <sub>72</sub> N <sub>2</sub> O <sub>12</sub> :	C, 60.94;	H, 9.69;	N, 3.74.
Found	C, 60.87;	H, 9.39;	N, 3.70.

35

Loss on drying at 120 °C, 0.74%.

40

45

50

55

## EXAMPLE 11

## Synthesis of 9-Deoxo-8a-aza-8a-methyl-8a-homoerythromycin A by Methylation of 9-Deoxo-8a-aza-8a-hydroxy-8a-homoerythromycin A 3'-N-Oxide

5

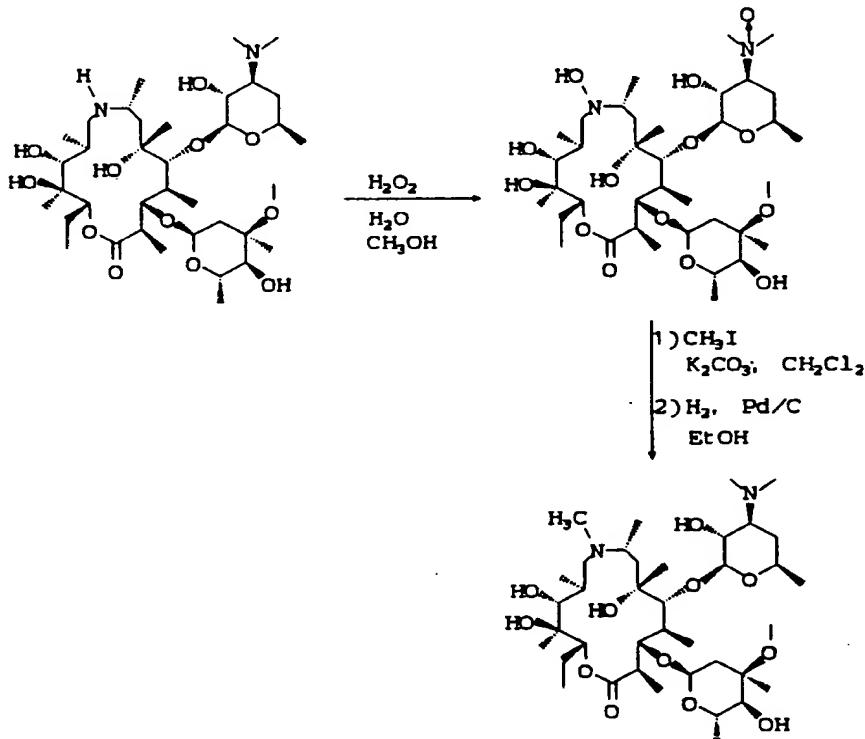
10

15

20

25

30



## Step 1

35

## 9-Deoxo-8a-aza-8a-hydroxy-8a-homoerythromycin A 3'-N-Oxide

9-Deoxo-8a-aza-8a-homoerythromycin A (385 mg, 0.524 mmol) in methanol (1.5 mL) was treated with 30% aqueous hydrogen peroxide (1.9 mL, 18.6 mmol) and the mixture was stirred at room temperature for 40 24 hours. The mixture was cooled in an ice bath, diluted with dichloromethane (10 mL) and water (8 mL), treated with saturated aqueous sodium sulfite (10 mL), and then stirred for 15 minutes to destroy excess 45 oxidant. The phases were separated and the aqueous portion extracted with more dichloromethane (2 x 15 mL). The combined organic solution was dried with magnesium sulfate, filtered, and evaporated under reduced pressure to afford crude 9-deoxo-8a-hydroxy-8a-aza-8a-homoerythromycin A 3'-N-oxide (349 mg) as a white solid.

## Step 2

50

## 9-Deoxo-8a-methyl-8a-aza-8a-homoerythromycin A

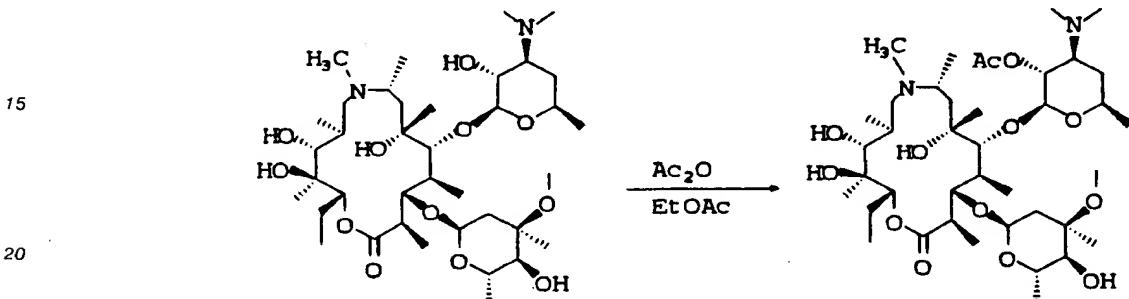
A portion of the product from step 1 (150 mg, 0.196 mmol) was dissolved in dichloromethane (3 mL) and the solution was treated with powdered, anhydrous potassium carbonate (2.0 g, 14.5 mmol) and methyl 55 iodide (0.5 mL, 8.0 mmol). The mixture was stirred at room temperature under a nitrogen atmosphere for 3.5 hours. The mixture was filtered and the solids washed with dichloromethane (5 mL). Water (3 mL) was added to the combined filtrate and washings and the mixture was stirred vigorously while the pH was brought to 11 with 1N sodium hydroxide. The dichloromethane phase was dried with magnesium sulfate, filtered and evaporated under reduced pressure to afford a mixture of 9-deoxo-8a-methyl-8a-homoerythromycin A 3'-N-oxide and 9-deoxo-8a-methyl-8a-homoerythromycin A 8a, 3'-N-bisoxide (136 mg) as a foam.

The crude product was dissolved in ethanol (6 mL), treated with 10% palladium on carbon (240 mg), and hydrogenated on a Parr shaker for 75 minutes at 45 psi. The mixture was filtered and the filtrate was evaporated under vacuum. The residue in dichloromethane (20 mL) was washed with saturated aqueous potassium carbonate, dried with magnesium sulfate, filtered, and evaporated under reduced pressure to 5 provide 9-deoxo-8a-methyl-8a-homoerythromycin A (107 mg) as a foam.

EXAMPLE 12

2'-O-Acetyl-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

10



25

To a solution of 1.5 g (2.0 mmol) of 9-deoxo-8a-aza-8a-methyl-8a homoerythromycin A in 30 ml of ethyl acetate at room temperature was added 0.306 g (2.99 mmol) of acetic anhydride and the mixture was stirred for 24 hours. To the reaction was then added 30 ml of water and the mixture was stirred for an additional 30 minutes. The pH of the aqueous layer was adjusted to the 2-3 range with 1N hydrochloric acid 30 and the organic and aqueous layer were separated. The pH of the aqueous layer was then adjusted to the 9-10 range with 1N sodium hydroxide solution and extracted with ethyl acetate. The ethyl acetate layer was washed with brine, dried (anhydrous sodium sulfate), and evaporated to yield 1.47 g (93% yield) of the title compound as a white foam.

TLC  $R_f$  0.37 ( $\text{CH}_2\text{Cl}_2\text{-MeOH-NH}_4\text{OH}$ , 9 : 1 : 0.1)

35 MP softens at 100-2°C, melts at 114-117°C

$^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  5.09 (d, H-1"), 4.81 (dd, H-13), 4.70 (dd, H-2"), 4.48 (br, H-3), 4.46 (d, H-1"), 3.30 (s,  $\text{OCH}_3$ ), 2.22 (s,  $\text{N}(\text{CH}_3)_2$ ), 2.03 (s,  $\text{OCOCH}_3$ ), 1.99 (s,  $\text{NCH}_3$ ).

$^{13}\text{C}$  NMR (400 MHZ,  $\text{CDCl}_3$ )  $\delta$  178.41, 169.89, 100.46, 94.17, 84.15, 78.16, 76.72, 75.95, 75.69, 74.75, 72.91, 71.49, 68.60, 66.53, 65.14, 63.69, 59.59, 56.41, 49.32, 45.26, 42.59, 40.66, 35.86, 34.70, 30.52, 30.28, 40 27.75, 21.98, 21.66, 21.48, 21.10, 18.14, 16.27, 14.21, 12.57, 11.89, 11.66, 11.38.

FAB Mass Spectrum (Li spike), m/z 797, 633, 574, 555.

45

50

55

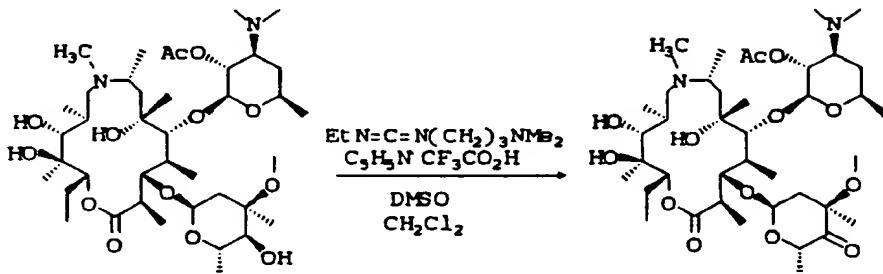
## EXAMPLE 13

2'-O-Acetyl-4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

5

10

15



To a solution of 5.9 g (7.4 mmol) of 2'-O-Acetyl-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A in 20 100 ml of dry methylene chloride under nitrogen at room temperature was successively added 5.3 ml (7.4 mmol) of dimethyl sulfoxide, 7.15 g (3.73 mmol) of 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride and 7.15 g (3.73 mmol) of pyridinium trifluoroacetate. The reaction mixture was stirred for 2 hours, at which point 300 ml of ethyl acetate followed by 100 ml of water was added. The pH of the solution was adjusted to the range of 9-10 with 1N sodium hydroxide solution. The ethyl acetate layer was washed 25 with brine, dried (anhydrous sodium sulfate) and evaporated. Silica chromatography with ethyl acetate-acetone-ammonium hydroxide (9 : 1 : 0.1) gave 4.8 g (82% yield) of the title compound as a white solid.

TLC  $R_f$  0.47 (EtOAc-Acetone-NH<sub>4</sub>OH, 10 : 1 : 0.1)

MP 68 -70°C

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  5.49 (t, H-1"), 4.85 (dd, H-13), 4.66 (dd, H-2'), 4.63 (br, H-3), 4.40 (q, H-5"), 4.36 (d, H-1'), 3.28 (s, OCH<sub>3</sub>), 2.20 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.07 (s, OCOCH<sub>3</sub>), 2.02 (s, NCH<sub>3</sub>).

<sup>13</sup>C NMR (400 MHz, CDCl<sub>3</sub>)  $\delta$  212.19, 178.62, 169.82, 99.77, 95.53, 83.81, 77.47, 77.34, 77.02, 74.69, 71.52, 71.25, 69.06, 66.55, 63.59, 59.89, 56.67, 51.31, 44.43, 40.59, 40.46, 36.74, 36.62, 30.74, 30.70, 30.50, 30.10, 29.22, 27.64, 22.00, 21.46, 21.40, 21.08, 16.23, 15.71, 13.75, 12.48, 11.89, 11.30.

FAB Mass Spectrum, m/z 789, 630, 599, 575, 551.

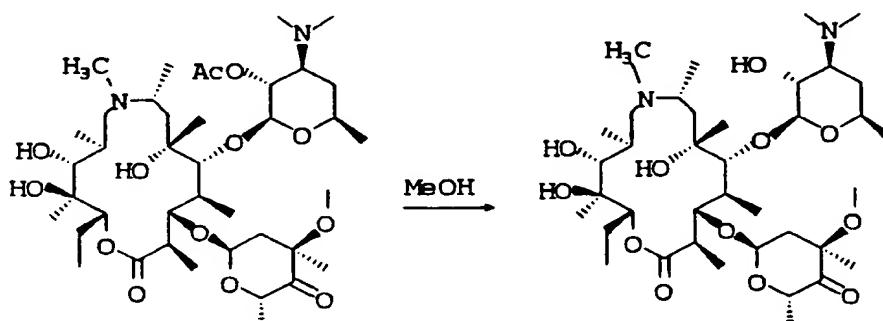
35

EXAMPLE 144"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

40

45

50



55 A solution of 4.8 g (6.1 mmol) of 2'-O-Acetyl-4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A in 160 ml of methanol was stirred at room temperature for 48 hours. Evaporation of the solvent gave 4.52 g (99% yield) of the title compound.

TLC  $R_f$  0.44 (CH<sub>2</sub>Cl<sub>2</sub>-MeOH-NH<sub>4</sub>OH, 9.3 : 0.7 : 0.1)

MP softens at 73°C, melts at 86-89°C

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 5.44 (t, H-1''), 4.82 (dd, H-13), 4.55 (br, H-3), 4.42 (q, H-5''), 4.22 (d, H-1'), 3.28 (s, OCH<sub>3</sub>), 3.15 (dd, H-2'), 2.24 (s, N(CH<sub>3</sub>)<sub>2</sub>), 2.00 (s, NCH<sub>3</sub>).

<sup>13</sup>C NMR (400 MHZ, CDCl<sub>3</sub>) δ 212.19, 178.89, 103.61, 95.32, 85.37, 77.47, 77.35, 77.13, 75.45, 74.88,

71.53, 70.37, 69.38, 66.30, 65.40, 59.50, 56.44, 51.29, 45.08, 42.45, 40.24, 36.63, 36.38, 36.30, 30.49, 28.50, 27.69, 21.90, 21.41, 21.30, 16.35, 15.85, 14.15, 12.45, 11.91, 11.83, 11.30.

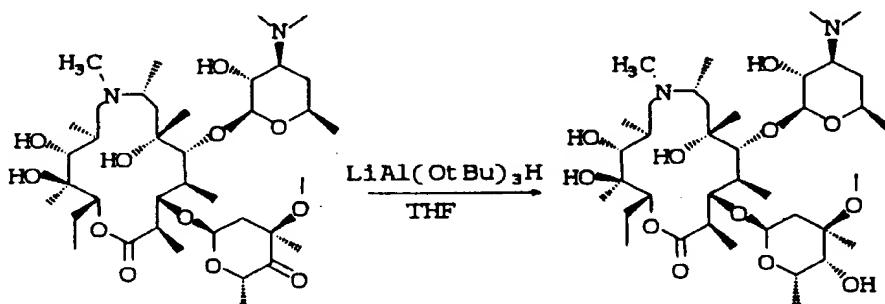
FAB Mass Spectrum (Li spike), m/z 753, 573, 398, 256.

EXAMPLE 15

10

4''-epi-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A

15



20

25

To 0.10 g (0.13 mmol) of 4''-deoxy-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A in 15 ml of dry tetrahydrofuran under nitrogen at room temperature was added 0.35 ml of lithium tri-tert-butoxy aluminohydride (1.0 M solution in THF). The reaction was judged to be complete by TLC after 15 minutes.

30 The reaction mixture was quenched with 5 ml of water followed by 30 ml of ethyl acetate. The pH of the solution was adjusted to the range of 9-10 with 1N sodium hydroxide solution. The ethyl acetate layer was separated and washed with brine, dried (anhydrous sodium sulfate), and evaporated. The mixture of 9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A and the 4''-epimer was separated by preparative TLC. The plate was eluted with CHCl<sub>3</sub>-acetone-NH<sub>4</sub>OH, 6 : 4 : 0.2 and was developed twice to yield 35 mg (35 %

35 yield) of the title compound.

TLC R<sub>f</sub> 0.37 (CHCl<sub>3</sub>-Acetone-NH<sub>4</sub>OH, 3 : 3 : 0.2)

<sup>1</sup>H NMR (400 MHz, CDCl<sub>3</sub>) δ 5.13 (br, H-1''), 4.77 (br, H-13), 4.55 (q, H-5''), 4.40 (br, H-3), 4.36 (d, H-1'), 3.27 (s, OCH<sub>3</sub>), 3.14 (dd, H-2'), 2.24 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.92 (s, NCH<sub>3</sub>).

FAB Mass Spectrum (Li spike), m/z 755, 591, 573, 159, 117.

40

45

50

55

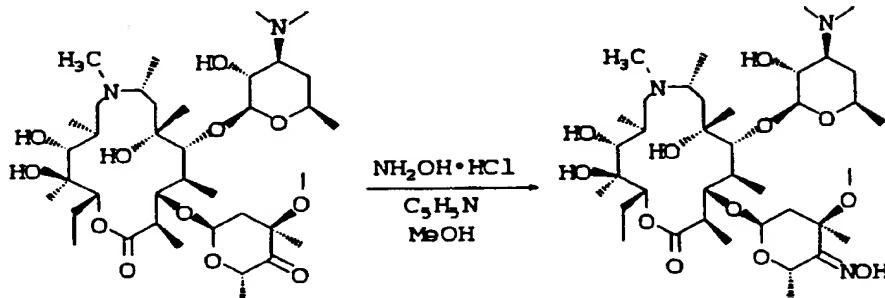
## EXAMPLE 16

4"-deoxy-4"-hydroxyimino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

5

10

15



20 To 2.5 g (3.3 mmol) of 4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A in 20 ml of methanol was added 2.7 ml (33 mmol) of pyridine followed by 1.2 g (16 mmol) of hydroxylamine hydrochloride. The resulting mixture was stirred for 48 hours at room temperature. Then the methanol was removed under vacuum and the residue was dissolved in 50 ml of water and 100 ml of ethyl acetate. The pH of the solution was adjusted to the range of 9-10 with 1N sodium hydroxide solution. The ethyl acetate layer was washed with brine, dried (anhydrous sodium sulfate), and evaporated. Silica chromatography with  $\text{CH}_2\text{Cl}_2$ -methanol- $\text{NH}_4\text{OH}$ , 93 : 7 : 1 afforded 1.5 g (58% yield) of the title compound as a mixtures of E and Z isomers.

25 TLC  $R_f$  0.37 ( $\text{CHCl}_3$ -Acetone- $\text{NH}_4\text{OH}$ , 3 : 3 : 0.2)

30 MP softens at 122°C, melts at 131-136°C

35  $^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  5.19 (t, H-1"), 5.06 (q, H-5"), 4.81 (dd, H-13), 4.38 (q, H-3), 4.23 (d, H-1'), 3.25 (s,  $\text{OCH}_3$ ), 3.17 (dd, H-2'), 2.23 (s,  $\text{N}(\text{CH}_3)_2$ ), 1.96 (s,  $\text{NCH}_3$ ).

40  $^{13}\text{C}$  NMR (400 MHz,  $\text{CDCl}_3$ )  $\delta$  178.58, 160.64, 103.16, 95.18, 84.95, 78.51, 77.35, 77.24, 77.04, 74.69, 70.66, 70.50, 69.38, 68.95, 66.62, 65.57, 65.40, 59.41, 56.24, 51.14, 50.45, 45.16, 42.03, 40.31, 39.45, 30.52, 28.95, 27.76, 25.27, 23.13, 21.87, 21.77, 21.33, 17.60, 16.98, 16.34, 14.84, 12.47, 12.05, 11.30, 11.05.

45 FAB Mass Spectrum (Li spike), m/z 768, 574, 416, 306

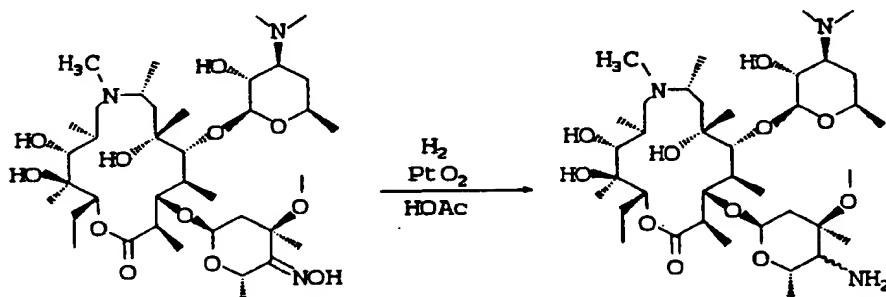
## EXAMPLE 17

4"-Deoxy-4"-amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

40

45

50



55 A solution of 0.50 g (0.65 mmol) of the oxime mixture (prepared as described in the previous example) in 5 ml of acetic acid was hydrogenated over 0.50 g of platinum oxide at 1000 psi at room temperature, initially for 48 hours. This was followed by a second addition of 0.20 g of platinum oxide and the reaction was continued for another 24 hours under 1000 psi. The reaction mixture was filtered and the acetic acid

was removed under vacuum. The residue was dissolved in 10 ml of water and 50 ml of ethyl acetate. The pH of the solution was adjusted to the range of 9-10 with 1N sodium hydroxide solution. The ethyl acetate layer was separated and washed with brine, dried (anhydrous sodium sulfate), and evaporated to give 0.47 g (95% yield) of the title compound. The product was shown by proton NMR to be a mixture of 4''-(S) ( $\delta$  4.61, q, H-5'') and 4''-(R) ( $\delta$  4.00, br, H-5'') amines, approximately in the ratio of 2 : 1.

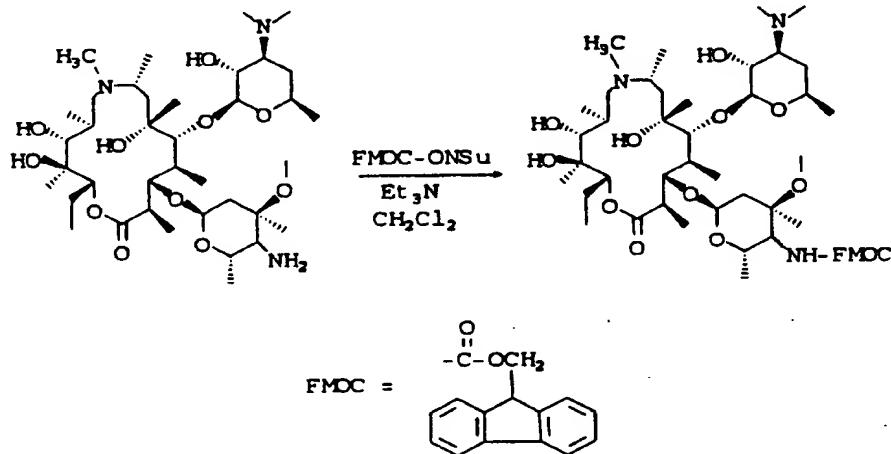
EXAMPLE 18

Separation of 4''-Deoxy-4''-(S) and 4''-(R)- amino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A

10

15

20



25

To a solution of 0.05 g (0.06 mmol) of the 4''-amino isomers, prepared as described in the previous example, in 1.5 ml methylene chloride at room temperature was added 0.027 g (0.080 mmol) of N-(9-fluorenylmethoxycarbonyloxy)succinimide (FMOC-ONSu) followed by 0.050 ml (0.33 mmol) of triethyl amine. The reaction mixture was stirred for 40 hours. The solvent was removed under vacuum and the residue was purified by preparative thin layer chromatography. The plate was developed with EtOAc-hexane-MeOH-NH<sub>4</sub>OH (8 : 2 : 0.15 : 0.15). Two compounds were isolated and the NMR analysis revealed that the less polar component ( $R_f$  = 0.48,  $\delta$  4.05, m, H-5'', 16.8 mg) was identified as an FMOC derivative of 4''-(S) amine (equatorial) and the slightly more polar component ( $R_f$  = 0.44,  $\delta$  4.66, q, H-5'', 6.6 mg) was identified as FMOC derivative of 4''-(R) amine (axial).

40

45

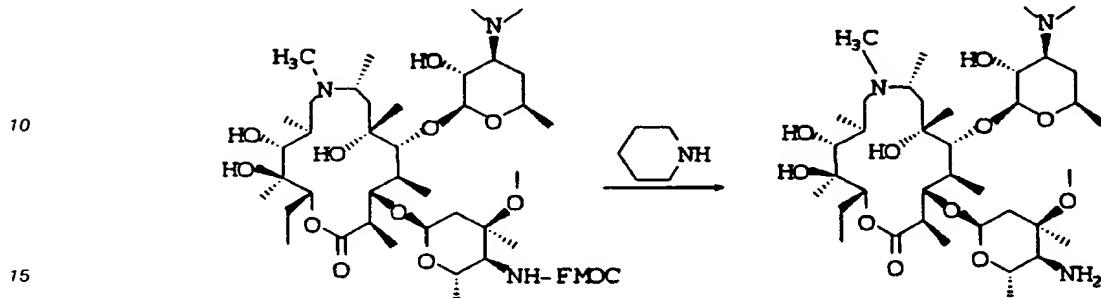
50

55

## EXAMPLE 19

FMOC deprotection to give 4"-Deoxy-4"--(S) - amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

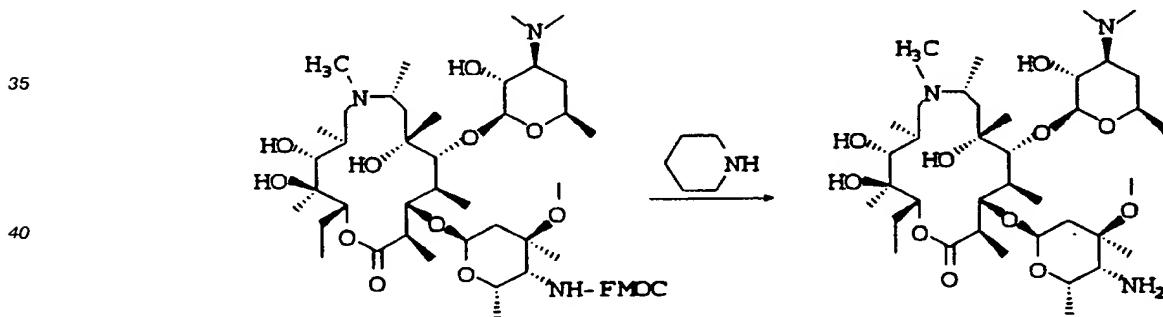
5



To 16.8 mg of the 4"--(S)-FMOC derivative was added 0.25 ml of piperidine and the mixture was stirred at room temperature. After 15 minutes TLC showed that the reaction was complete at which point piperidine was removed under vacuum and the residue purified by silica chromatography. Elution with  $\text{CH}_2\text{Cl}_2$ -MeOH- $\text{NH}_4\text{OH}$ , 9.3 : 0.7 : 0.1 gave 8.2 mg of 4"-deoxy-4"- (S) - amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A as a white solid.

$^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ , at 60°C)  $\delta$  5.10 ( d, H-1"), 4.82 (dd, H-13), 4.46 (q, H-3), 4.39 (d, H-1'), 4.00 (br, H-5"), 3.27 (s,  $\text{OCH}_3$ ), 2.38 (s,  $\text{N}(\text{CH}_3)_2$ ), 2.03 (s,  $\text{NCH}_3$ ).

## EXAMPLE 20

FMOC deprotection to give 4"-Deoxy-4"--(R) - amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

To 7.0 mg of the 4"--(R)-FMOC derivative was added 0.25 ml of piperidine and the mixture was stirred at room temperature. After 15 minutes TLC showed that the reaction was complete at which point piperidine was removed under vacuum and the residue purified by silica chromatography. Elution with  $\text{CH}_2\text{Cl}_2$ -MeOH- $\text{NH}_4\text{OH}$ , 9.3 : 0.7 : 0.1 gave 4.3 mg of 4"-deoxy-4"- (R) - amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A as a white solid.

$^1\text{H}$  NMR (400 MHz,  $\text{CDCl}_3$ , at 60°C)  $\delta$  5.10 ( d, H-1"), 4.82 (dd, H-13), 4.61 (q, H-5"), 4.48 (q, H-3), 4.41 (d, H-1'), 3.30 (s,  $\text{OCH}_3$ ), 3.16 (dd, H-2'), 2.27 (s,  $\text{N}(\text{CH}_3)_2$ ), 2.01 (s,  $\text{NCH}_3$ ).

55

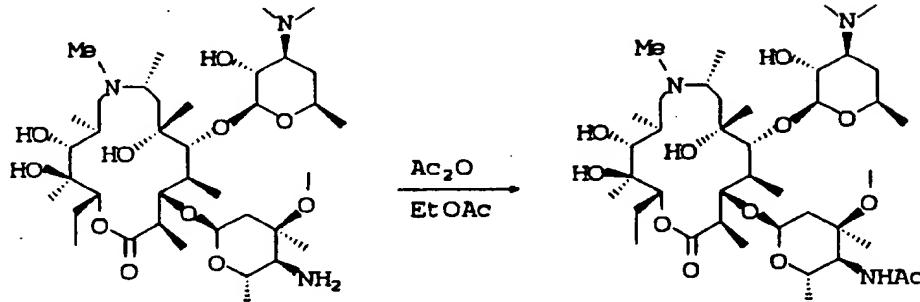
## EXAMPLE 21

## 4"-Deoxy-4"-(S)-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin-A

5

10

15



20

To a solution of 17.0 mg (0.022 mmol) of 4"-deoxy-4"-(S)-amino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A in 1.0 ml ethyl acetate was added 4.7 mg (0.046 mmol) of acetic anhydride and the mixture was stirred for 24 hours. To the reaction was then added 1.0 ml of water and the mixture was stirred for an additional 30 minutes. The pH of the aqueous layer was then adjusted to the 9-10 range with

25 1N sodium hydroxide solution and this layer was extracted with ethyl acetate. The ethyl acetate layer was washed with brine, dried (anhydrous sodium sulfate), and evaporated to yield crude product that was identified by NMR to be 2'-O-acetyl-4"-Deoxy-4"-(S)-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin-A (diagnostic signals at  $\delta$  4.72 dd, H'2' and at 3.70, t, H-4"). This product was not purified but was directly solvolysed by simply stirring in methanol at room temperature for 24 hours.

30 Evaporation of methanol followed by silica chromatography and elution with  $\text{CH}_2\text{Cl}_2$ -MeOH-NH<sub>4</sub>OH, 95:5:1 gave 16.2 mg of 4"-deoxy-4"-(S)-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A as a white solid.

TLC  $R_f$  0.50 ( $\text{CH}_2\text{Cl}_2$ -MeOH-NH<sub>4</sub>OH, 9.5:0.5:0.1)

MP 131 °-135 °C

35 <sup>1</sup>H NMR (400 MHz, at 60 °C,  $\text{CDCl}_3$ )  $\delta$  5.76 (d, NHAc), 5.13 (d, H-1"), 4.83 (dd, H-13), 4.49 (t, H-3), 4.30 (d, H-1'), 4.07 (m, H-5"), 3.70 (t, H-4"), 3.25 (s,  $\text{OCH}_3$ ), 2.29 (br,  $\text{N}(\text{CH}_3)_2$ ), 3.01 (s,  $\text{NHCOCH}_3$ ), 1.96 (s,  $\text{NCH}_3$ ). FAB mass spectrum (Li spike), m/z 796, 579, 374, 306, 161.

40

45

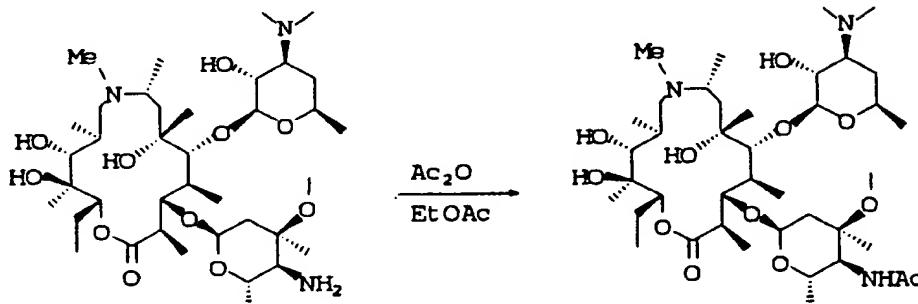
50

55

## EXAMPLE 22

4"-Deoxy-4"-*(R)*-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A

5

10  
15

20 To a solution of 20.0 mg (0.026 mmol) of 4'-deoxy-4"-*(R)*-amino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A in 1.0 ml ethyl acetate was added 5.8 mg (0.056 mmol) of acetic anhydride and the mixture was stirred for 24 hours. To the reaction was then added 1.0 ml of water and the mixture was stirred for an additional 30 minutes. The pH of the aqueous layer was then adjusted to the 9-10 range with 1N sodium hydroxide solution and this layer was extracted with ethyl acetate. The ethyl acetate layer was washed with brine, dried (anhydrous sodium sulfate), and evaporated to yield crude product that was identified by NMR to be 2'-O-acetyl-4"-*(R)*-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin-A (diagnostic signals at  $\delta$  4.70 dd, H'2' and at 3.66, d, H-4"). This product was not purified but was directly solvolyzed by simply stirring in methanol at room temperature for 24 hours. Evaporation of methanol followed by silica chromatography and elution with  $\text{CH}_2\text{Cl}_2$ -MeOH-NH<sub>4</sub>OH, 95:5:1 gave 17.6 mg of 4'-deoxy-4"-*(R)*-acetylamino-9-deoxy-8a-aza-8a-methyl-8a-homoerythromycin A as a white solid.

30 TLC  $R_f$  0.60 ( $\text{CH}_2\text{Cl}_2$ -MeOH-NH<sub>4</sub>OH, 9.0:1.0:0.1)  
MP 153°-156° C  
<sup>1</sup>H NMR (400 MHz, at 60 °C,  $\text{CDCl}_3$ )  $\delta$  5.75 (d, NHAc), 5.17 (d, H-1"), 4.83 (dd, H-13), 4.68 (q, H-5), 4.48 (br, H-3'), 4.39 (d, H-1"), 3.74 (d, H-4"), 3.29 (s,  $\text{OCH}_3$ ), 2.27 (br,  $\text{N}(\text{CH}_3)_2$ ), 2.01 (s,  $\text{NHCOCH}_3$ ), 1.99 (s,  $\text{NCH}_3$ ).

35 FAB mass spectrum (Li spike), m/z 796, 597, 579, 366, 306.

40

45

50

55

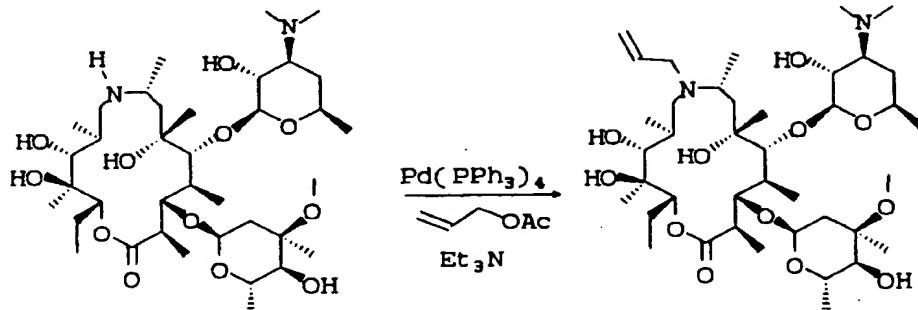
## EXAMPLE 23

## 9-Deoxo-8a-aza-8a-allyl-8a-homoerythromycin A

5

10

15



20 A mixture of 9-deoxo-8a-aza-8a-homoerythromycin A (5.0 g, 6.8 mmol), allyl acetate (35 mL), triethylamine (2.5 mL) and tetrakis(triphenylphosphine)palladium(0) (0.23 g) was stirred and heated in an oil bath at 80 °C for 7 hours. After stirring at room temperature overnight, the mixture was evaporated under vacuum to remove excess allyl acetate. The residue was stirred with dichloromethane (50 mL) and water (50 mL) while the pH was adjusted from 5.6 to 4.0 with 2N hydrochloric acid. The layers were separated  
 25 and the organic portion was extracted with more water (15 mL). The combined aqueous solution was layered with dichloromethane and stirred while the pH was brought to 9.4 by addition of 5N aqueous sodium hydroxide solution. The layers were separated and the aqueous portion was extracted with additional dichloromethane (50 mL, 25 mL). The combined dichloromethane extracts were dried over magnesium sulfate, filtered and evaporated under vacuum to a foam (4.85 g).

30 A solution of the foam in 2-propanol (40 mL) deposited crystals on stirring at room temperature. The crystalline product was collected, washed with 2-propanol (3 x 4 mL) and dried under a nitrogen stream to afford 9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (1.8 g) as a white solid. The mother liquors and washings were evaporated under vacuum and the residue stirred with ethanol (12 mL) to afford a second crop (1.5 g) of the title compound.

35  $^1\text{H}$  NMR ( $\text{CDCl}_3$ )  $\delta$  5.90 (m,  $\text{CH}=\text{CH}_2$ ), 5.13 (d,  $\text{H}=\text{CHaHb}$ ), 5.03 (m,  $\text{CH}=\text{CHaHb}$  and  $\text{H-1''}$ ), 4.92 (m,  $\text{H-13}$ ), 4.36 (m,  $\text{H-3}$ ), 4.35 (d,  $\text{H-1'}$ ), 4.03 (m,  $\text{H-5''}$ ), 3.62 (br s,  $\text{H-11}$ ), 3.50 (m,  $\text{H-5}$  and  $\text{H-5'}$ ), 3.27 (s,  $\text{OCH}_3$ ), 3.24 (m,  $\text{H-2'}$ ), 2.26 (s,  $\text{N}(\text{CH}_3)_2$ ), 1.87 (m,  $\text{H-14a}$ ), 1.65 (m,  $\text{H-4'eq}$ ), 1.53 (dd,  $\text{H-2'ax}$ ), 1.44 (m,  $\text{H-14b}$ ), 1.29 (br s,  $6\text{-CH}_3$ ), 0.87 (t,  $\text{CH}_2\text{CH}_3$ ).

FAB-MS m/z 798 ( $\text{M} + \text{Na}$ ), 776 ( $\text{M} + \text{H}$ ), 618, 600, 159.

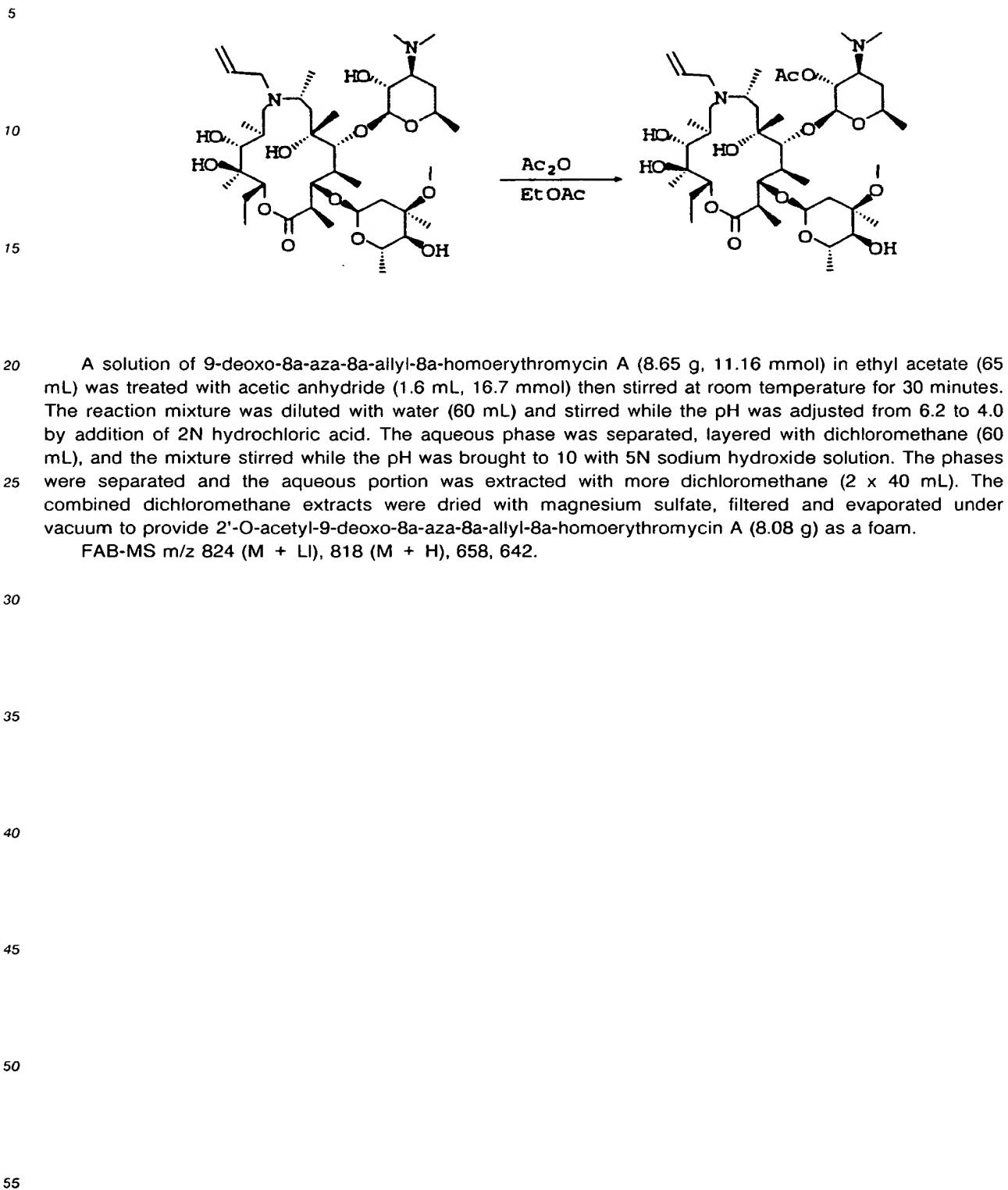
40

45

50

55

## EXAMPLE 24

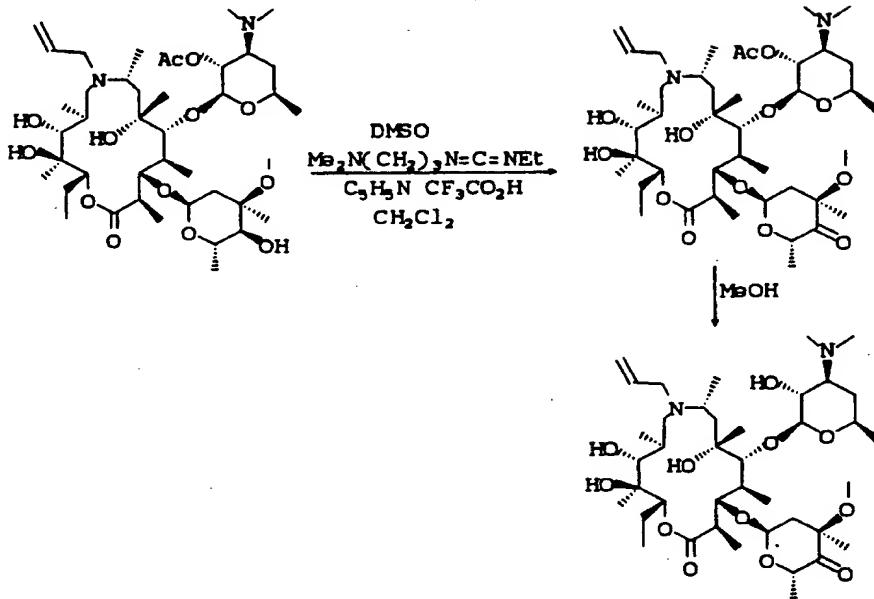
2'-O-Acetyl-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A

## EXAMPLE 25

## 4'-Deoxy-4'-oxo-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A

5

10



15

20

25

A solution of 2'-O-acetyl-9-deoxy-8a-aza-8a-allyl-8a-homoerythromycin A (8.0 g, 9.79 mmol) in dichloromethane (75 mL) and dimethylsulfoxide (7 mL, 97.9 mmol) was treated over two minutes with 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (5.64 g, 29.4 mmol) and then over three minutes with pyridinium trifluoroacetate (5.7 g, 29.4 mmol). The resulting mixture was stirred at room temperature for 2.5 hours, then diluted with water (75 mL) and stirred vigorously while the pH of the aqueous phase was adjusted from 4.6 to 9.5 by addition of 5N sodium hydroxide solution. The phases were separated and the aqueous portion was extracted with more dichloromethane (2 x 50 mL). The combined dichloromethane extracts were washed with aqueous 5% sodium bicarbonate (3 x 50 mL), dried over magnesium sulfate, filtered and evaporated under vacuum to afford crude 2'-O-acetyl-4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (8.4 g) as a foam.

The crude product was dissolved in methanol (50 mL) and the solution was stirred at room temperature overnight followed by one hour at 50 °C. After cooling to room temperature, the mixture was evaporated under vacuum to afford 4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (7.7 g) as a foam.

FAB-MS m/z 780 (M + Li), 774 (M + H), 616, 600.

45

50

55

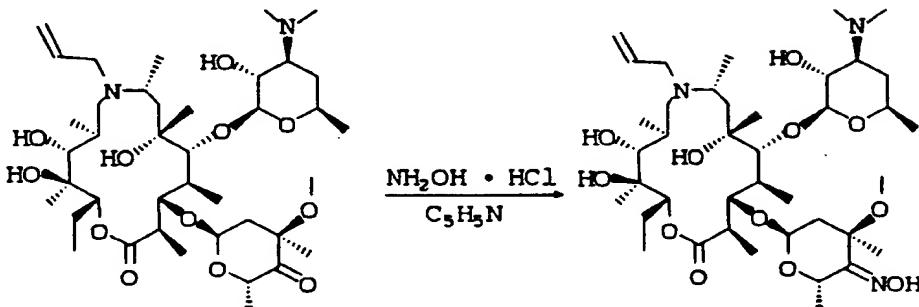
## EXAMPLE 26

## 4"-Deoxy-4"-hydroxyimino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A

5

10

15



20 Hydroxylamine hydrochloride (3.4 g, 49 mmol) was added to a solution of 4"-deoxy-4"-oxo-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (4.0 g, 4.9 mmol) in pyridine (12 mL). The resulting mixture was stirred at room temperature for 16 hours, then evaporated under vacuum to remove the pyridine. The residue was stirred with dichloromethane (100 mL) and water (100 mL, pH of aqueous phase was 5.9) and the layers separated. The aqueous portion was stirred with additional dichloromethane (100 mL) while adjusting the pH to 6.0 with 5N sodium hydroxide solution. The extraction process was repeated with three additional 100 mL portions of dichloromethane with adjustment of the pH of the aqueous phase to values of 6.5, 7.0 and 11.0. The pH 7.0 and 11.0 extracts were combined and evaporated under vacuum to provide 4"-deoxy-4"-hydroxyimino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (2.36 g) as a mixture of oxime isomers.

30  $^1\text{H}$  NMR ( $\text{CDCl}_3$ , 60 °C)  $\delta$  5.95 (br m,  $\text{CH}=\text{CH}_2$ ), 5.32 (t, H-1''), 5.14 (m,  $\text{CH}=\text{CH}_2$ ), 5.05 (m, H-5''), 4.92 (dd, H-13), 4.42 (d, H-1''), 4.38 (m, H-3), 3.63 (br s, H-11), 3.31 (s,  $\text{OCH}_3$ ), 2.48 (br s,  $\text{N}(\text{CH}_3)_2$ ), 0.90 (t,  $\text{CH}_2\text{CH}_3$ ).

FAB-MS m/z 795 (M + Li), 789 (M + H), 630, 616, 600.

## EXAMPLE 27

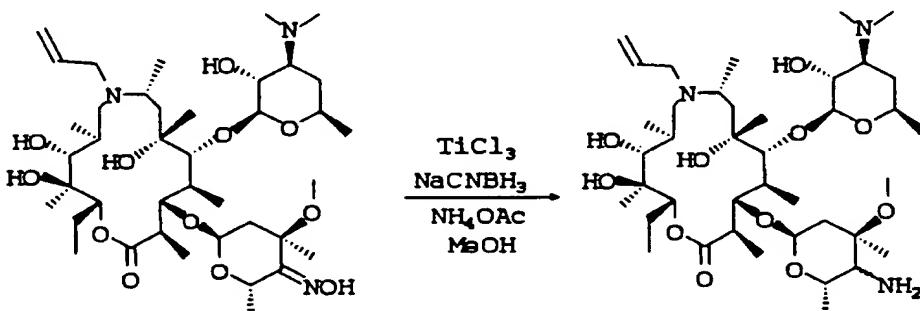
35

## 4"-Deoxy-4"-amino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A

40

45

50



55 A solution of 4"-deoxy-4"-hydroxyimino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (2.0 g, 2.4 mmol) in methanol (50 mL) was treated sequentially with ammonium acetate (4.5 g, 58 mmol), sodium cyanoborohydride (0.5 g, 95% pure, 7.6 mmol), and titanium (III)chloride (5.0 mL of a 15% solution in 20% hydrochloric acid, 5.8 mmol). The resulting mixture was stirred at room temperature for 27 hours, then treated with nitromethane (40 drops) and stirred an additional 15 minutes to reduce excess oxidant. The apparent pH of the mixture was adjusted from 6.05 to 7.0 with 5N sodium hydroxide before evaporating the

solvent under vacuum. The residue was stirred with dichloromethane (200 mL) and water (200 mL) while adjusting the pH of the aqueous portion to 9.5 with 5N sodium hydroxide. The layers were separated and the aqueous portion extracted with additional dichloromethane (2 x 100 mL). The combined dichloromethane extracts were dried over magnesium sulfate, filtered and evaporated under vacuum to afford the crude 4"-amino derivative (1.88 g).

A portion (1.6 g) of the crude product was purified by column chromatography on EM silica gel 60 (4.5 x 23 cm, 230-400 mesh, packed under 90:10:1 dichloromethane-methanol-conc. ammonium hydroxide). The column was eluted with 90:10:1 dichloromethane-methanol-conc. ammonium hydroxide, collecting 25 mL fractions. Fractions 15-24 were combined, evaporated under vacuum, and the residue lyophilized from benzene to afford 4"-deoxy-4"-amino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (1.02 g) as a 3:2 (4"R):(4"S) mixture of isomers.

<sup>1</sup>H NMR (CDCl<sub>3</sub>, 60 °C) δ 5.94 (m, CH = CH<sub>2</sub>), 5.08 (m, CH = CH<sub>2</sub> and H-1"), 4.93 (m, H-13), 4.64 (q, H-5" of the 4"R-isomer), 4.39 (m, H-1' and H-3), 4.06 (dq, H-5" of the 4"S-isomer), 3.65 (s, H-11), 3.54 (m, H-5 and H-5'), 3.28 (s, OCH<sub>3</sub> of the 4"R-isomer), 3.26 (s, OCH<sub>3</sub> of the 4"S-isomer), 2.36 (br s, N(CH<sub>3</sub>)<sub>2</sub>).

FAB-MS m/z 781 (M + Li), 775 (M + H), 617, 616, 600.

#### EXAMPLE 28

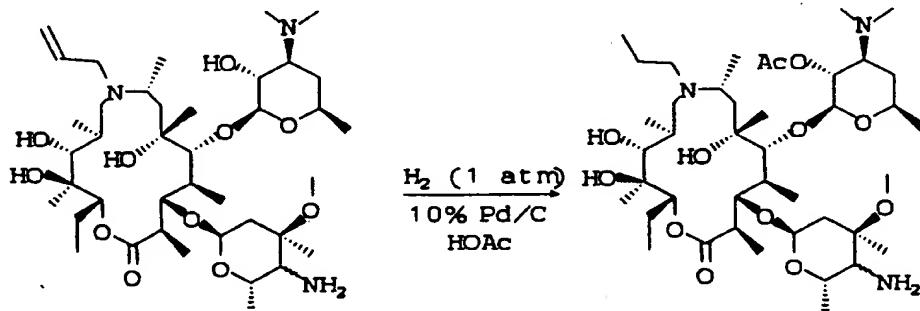
##### 4"-Deoxy-4"-amino-9-deoxo-8a-aza-8a-propyl-8a-homoerythromycin A

20

25

30

35



40

A mixture of 4"-deoxy-4"-amino-9-deoxo-8a-aza-8a-allyl-8a-homoerythromycin A (ca. 150 mg of a mixture of 4"-isomers), 10% palladium on carbon (120 mg) and acetic acid (5 mL) was stirred under a hydrogen atmosphere at room temperature for 5 hours. The mixture was filtered and the filtrate evaporated under vacuum. The residue was stirred with dichloromethane (6 mL) and water (4 mL) while adjusting the pH to 10 with dilute aqueous sodium hydroxide. The layers were separated and the aqueous portion was extracted with more dichloromethane (4 mL). The combined extracts were dried over magnesium sulfate, filtered and evaporated under vacuum to provide crude product.

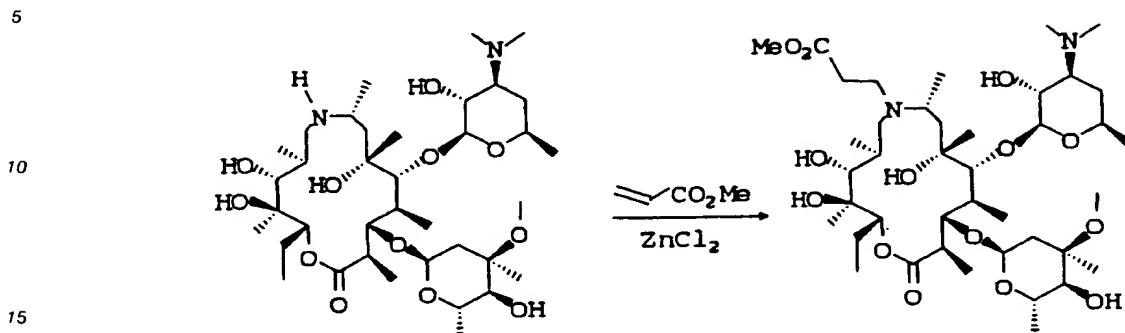
The crude product was purified by chromatography on a column of EM silica gel 60 (4.5 x 14 cm, 230-400 mesh). The column was packed and eluted with 90:10:1 dichloromethane-methanol-conc. ammonium hydroxide, collecting 8 mL fractions. Fractions 25-30 were combined and evaporated under vacuum to provide 4"-deoxy-4"-amino-9-deoxo-8a-aza-8a-propyl-8a-homoerythromycin A (121 mg) as a mixture of 4"-isomers (ca. 2:1 R:S).

<sup>1</sup>H NMR (CDCl<sub>3</sub>, 60 °C) δ 4.66 (q, H-5" of the 4"R-isomer), 4.03 (m, H-5" of the 4"S-isomer), 3.27 (s, OCH<sub>3</sub> of the 4"R-isomer), 3.25 (s, OCH<sub>3</sub> of the 4"S-isomer), 2.40 (br s, N(CH<sub>3</sub>)<sub>2</sub>).

FAB-MS m/z 777 (M + H), 619, 618, 602, 158.

55

## EXAMPLE 29

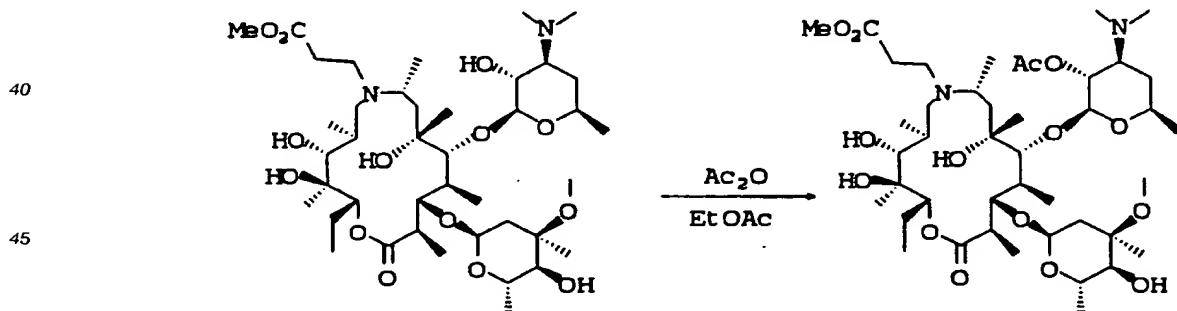
9-Deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A

20 Zinc chloride (215 mg, 1.58 mmol) and methanol (0.11 mL, 2.7 mmol) were added to a mixture of 9-deoxy-8a-aza-8a-homoerythromycin A (2.0 g, 2.7 mmol) and methyl acrylate (30 mL, 333 mmol). The resulting mixture was stirred at 60 °C for 91 hours. The excess methyl acrylate was removed under vacuum leaving a white solid residue. The crude product was purified by flash chromatography on EM silica gel 60 (7.0 x 30.5 cm, column eluted with 95:5:1 chloroform: methanol:aqueous ammonia collecting 50 mL fractions). Fractions 29-40 were combined and evaporated under vacuum to afford 9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (1.58 g) as a foam.

25 <sup>1</sup>H NMR (400 MHz, CD<sub>3</sub>OD) δ 5.14 (d, H-1''), 4.97 (dd, H-13), 4.42 (br s, H-3), 4.39 (d, H-1'), 4.19 (m, H-5''), 3.67 (s, CO<sub>2</sub>CH<sub>3</sub>), 3.64 (m, H-5'), 3.51 (d, H-5), 3.48 (br s, H-11), 3.31 (s, OCH<sub>3</sub>), 3.27 (dd, H-2'), 3.00 (d, H-4''), 2.36 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.34 (br s, 6-CH<sub>3</sub>), 1.14 (d, 8-CH<sub>3</sub>), 1.10 (s, 12-CH<sub>3</sub>), 1.02 (d, 4-CH<sub>3</sub>), 0.93 (d, 10-CH<sub>3</sub>), 0.89 (t, CH<sub>2</sub>CH<sub>3</sub>).

30 FAB-MS, m/z 821 (M + H), 827 (M + Li).

## EXAMPLE 30

2'O-Acetyl-9-deoxy-8a-aza-8a-methoxycarbonethyl-8a-homoerythromycin A

50 Acetic anhydride (0.062 mL, 0.66 mmol) was added to a solution of 9-deoxy-8a-aza-8a-(3-methoxy-3-oxo-propyl)-8a-homoerythromycin A (355 mg, 0.43 mmol) in dry ethyl acetate (2.7 mL). The resulting solution was stirred at room temperature for 3.5 hours. Water (2 mL) was then added and the two-phase mixture was stirred at room temperature for 2 hours. The pH of the aqueous phase was adjusted to 2.5 by addition of 1N hydrochloric acid. The layers were separated and the organic phase was discarded. Fresh ethyl acetate was added and the pH of the aqueous layer was adjusted to 9.5 by addition of 1N sodium hydroxide. The layers were separated and the aqueous layer was extracted three times with ethyl acetate. The combined ethyl acetate extracts were dried over magnesium sulfate, filtered and evaporated under

vacuum to afford 2'-O-acetyl-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (299 mg) as a clear colorless oil. The crude product was essentially pure and was used without further purification in the next step.

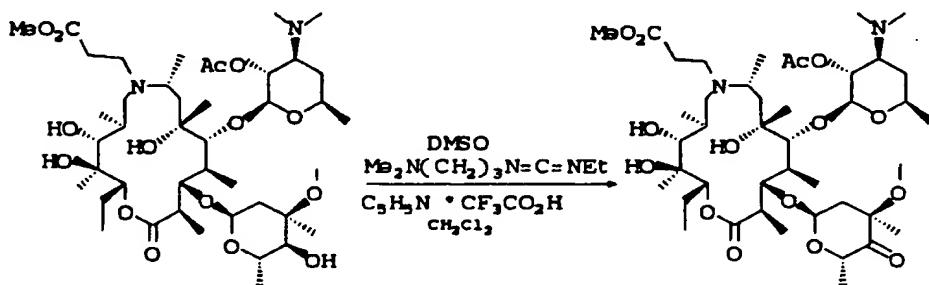
5  $^1\text{H}$  NMR (400 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  5.11 (br s, H-1''), 4.75 (dd, H-2'), 4.65 (d, H-1'), 4.29 (br s, H-3), 4.15 (m, H-5''), 3.71 (m, H-5'), 3.67 (s,  $\text{CO}_2\text{CH}_3$ ), 3.56 (d, H-5), 3.48 (br s, H-11), 3.35 (s,  $\text{OCH}_3$ ), 3.03 (d, H-4''), 2.27 (s,  $\text{N}(\text{CH}_3)_2$ ), 2.08 (s,  $\text{OCOCH}_3$ ), 1.33 (br s, 6- $\text{CH}_3$ ), 1.11 (s, 12- $\text{CH}_3$ ), 0.91 (t,  $\text{CH}_2\text{CH}_3$ ).

EXAMPLE 31

10

15

20



25 1-(3-Dimethylaminopropyl)-3-ethylcarbodiimide hydrochloride (200 mg, 1.04 mmol) was added to a solution of 2'-O-acetyl-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (299 mg, 0.35 mmol) in dry dichloromethane (2.1 mL). Methyl sulfoxide (0.246 mL, 3.5 mmol) was then added dropwise followed by dropwise addition of a solution of pyridinium trifluoroacetate (204 mg, 1.06 ml mmol) in dry dichloromethane (0.7 mL). The reaction mixture was then stirred at room temperature for 110 minutes. Aqueous potassium carbonate (5%) was added and the pH of the aqueous layer was adjusted to 10 by addition of solid potassium carbonate. The layers were separated and the aqueous layer was extracted four times with ethyl acetate. The combined organic layers were dried over magnesium sulfate, filtered and evaporated under vacuum to afford 2'-O-acetyl-4'-deoxy-4'-oxo-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A as a clear yellow oil (746 mg). The crude product contains some residual DMSO and other reagent derived impurities. However, it was used without further purification in the next step.

30 35  $^1\text{H}$  NMR (400 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  5.56 (m, H-1''), 5.03 (dd, H-13), 4.79 (dd, H-2'), 4.58 (br s, H-3), 4.56 (d, H-1'), 4.53 (q, H-5''), 3.66 (s,  $\text{CO}_2\text{CH}_3$ ), 3.58 (d, H-5), 3.55 (m, H-5'), 3.41 (br s, H-11), 3.26 (s,  $\text{OCH}_3$ ), 2.26 (s,  $\text{N}(\text{CH}_3)_2$ ), 2.10 (s,  $\text{OCOCH}_3$ ), 0.90 (t,  $\text{CH}_2\text{CH}_3$ ).

40

45

50

55

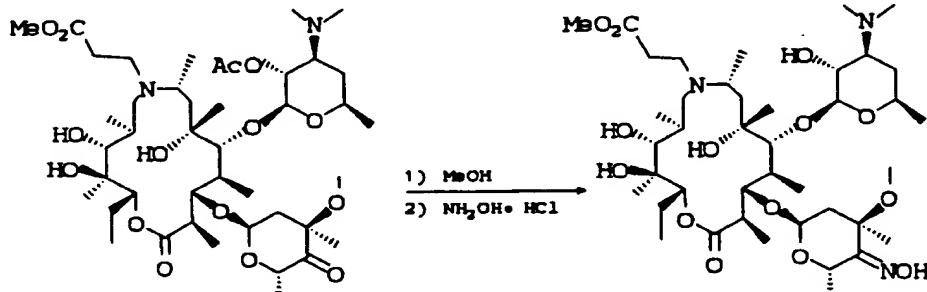
## EXAMPLE 32

## 4"-Deoxy-4"-hydroxyimino-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A

5

10

15



A solution of crude 2'O-acetyl-4"-deoxy-4"-oxo-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (746 mg) in methanol (4 mL) was stirred at room temperature for 19 hours. Analytical TLC showed complete deacetylation. Hydroxylamine hydrochloride (121 mg) was then added. The reaction mixture was stirred at room temperature for 1.5 hours then evaporated under vacuum. The residue was partitioned between ethyl acetate and aqueous potassium carbonate (pH of aqueous layer was adjusted to 10 by addition of solid potassium carbonate). The aqueous layer was extracted four times with ethyl acetate and the combined organic layers were dried over magnesium sulfate, filtered and evaporated under vacuum to afford a clear colorless oil (294 mg). The crude product was purified by flash chromatography on EM silica gel 60 (4.0 x 48 cm column eluted with 95:5:1 chloroform:methanol:aqueous ammonia collecting 30 mL fractions). Fractions 23-34 were combined and evaporated under vacuum to afford 4"-deoxy-4"-hydroxyimino-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (169 mg) as a foam.

<sup>1</sup>H NMR (400 MHz, CD<sub>3</sub>OD) δ 5.33 (dd, H-1"), 5.16 (q, H-5"), 5.05 (dd, H-13), 4.42 (d, H-1'), 4.31 (br s, H-3), 3.71 (br s, H-11), 3.66 (s, CO<sub>2</sub>CH<sub>3</sub>), 3.65 (d, H-5), 3.58 (m, H-5'), 3.28 (s, OCH<sub>3</sub>), 3.25 (dd, H-2'), 2.32 (s, N-(CH<sub>3</sub>)<sub>2</sub>), 1.31 (br s, 6-CH<sub>3</sub>), 1.10 (s, 12-CH<sub>3</sub>), 1.08 (d, 8-CH<sub>3</sub>), 1.03 (d, 4-CH<sub>3</sub>), 0.96 (d, 10-CH<sub>3</sub>), 0.90 (t, CH<sub>2</sub>CH<sub>3</sub>).

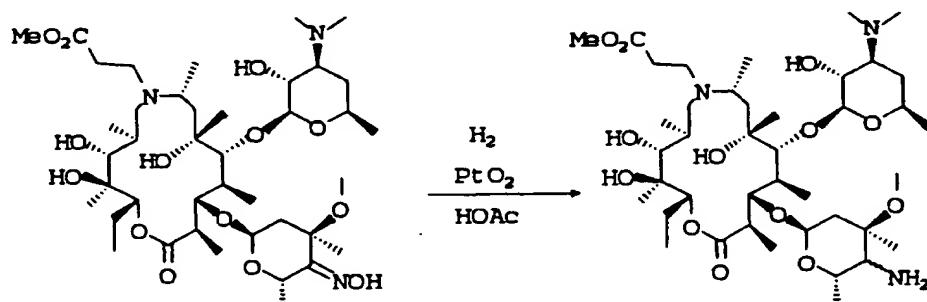
## EXAMPLE 33

## 4"-Deoxy-4"-amino-9-deoxy-8a-aza-8a-(3-methoxy-3-oxo-propyl)-8a-homoerythromycin A

40

45

50



A solution of 4"-deoxy-4"-hydroxyimino-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (169 mg, 0.19 mmol) in absolute ethanol was stirred with Raney nickel for 2 hours at room temperature to remove potential catalyst poisons. The Raney nickel was removed by filtration and the filtrate was evaporated under vacuum. The residue was dissolved in acetic acid (4 mL) then platinum oxide (123 mg, 0.54 mmol) was added. The reaction mixture was shaken under a hydrogen atmosphere (1000 psi) at room temperature for 48 hours. The mixture was then filtered through Celite® and the filtrate was

evaporated under vacuum. The residue was partitioned between 10% aqueous sodium bicarbonate and ethyl acetate. The aqueous layer was extracted six times with ethyl acetate. The combined organic layers were dried over magnesium sulfate, filtered and evaporated under vacuum to afford a colorless oil (116 mg). Analytical TLC indicated that no reaction had occurred. The crude material was therefore redissolved in 5 acetic acid (4 mL) and platinum oxide (115 mg, 0.50 mmol, different batch) was added. The mixture was shaken under an atmosphere of hydrogen (1000 psi) at room temperature for 72 hours. Workup as before afforded 4"-deoxy-4"-amino-9-deoxy-8a-aza-8a-(3-methoxy-3-oxopropyl)-8a-homoerythromycin A (ca. 1:1 mixture of epimers at C-4") as a white solid (73 mg).

10  $^1\text{H}$  NMR (400 MHz,  $\text{CD}_3\text{OD}$ )  $\delta$  5.23 and 5.19 (two s,  $\text{N}(\text{CH}_3)_2$ ).

15 FAB-MS,  $m/z$  826 ( $\text{M} + \text{Li}$ ).

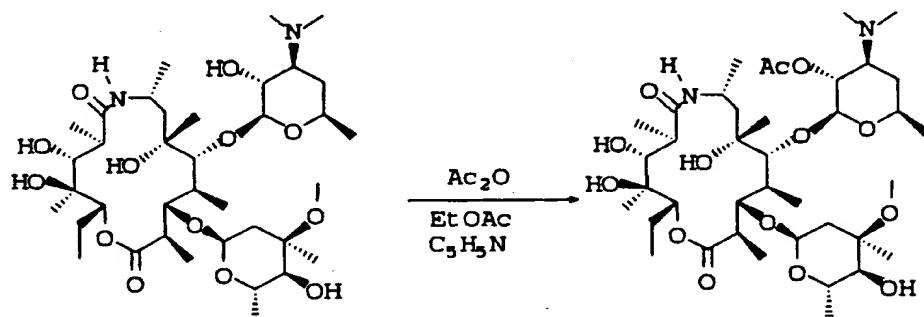
## EXAMPLE 34

2'-O-Acetyl-8a-aza-8a-homoerythromycin

15

20

25



30 A solution of 8a-aza-8a-homoerythromycin A (1.37 g, 1.83 mmol) and acetic anhydride (0.26 mL, 2.75 mmol) in ethyl acetate (12 mL) was stirred at room temperature for 27 minutes, then treated with pyridine (0.15 mL, 1.83 mmol) and stirred an additional 18 minutes. The mixture was evaporated under vacuum to remove most of the ethyl acetate. The residue was stirred with dichloromethane (12 mL) and water (12 mL) while the pH of the aqueous phase was carefully adjusted to 11 by addition of aqueous sodium hydroxide.

35 The layers were separated and the aqueous portion was extracted with more dichloromethane (12 mL). The combined extracts were dried with magnesium sulfate, filtered and evaporated under vacuum to provide 2'-O-acetyl-8a-aza-8a-homoerythromycin A (1.45 g) as a foam. This material was used in the next step without further purification.

40

45

50

55

## EXAMPLE 35

4"-Deoxy-4"-oxo-8a-aza-8a-homoerythromycin A

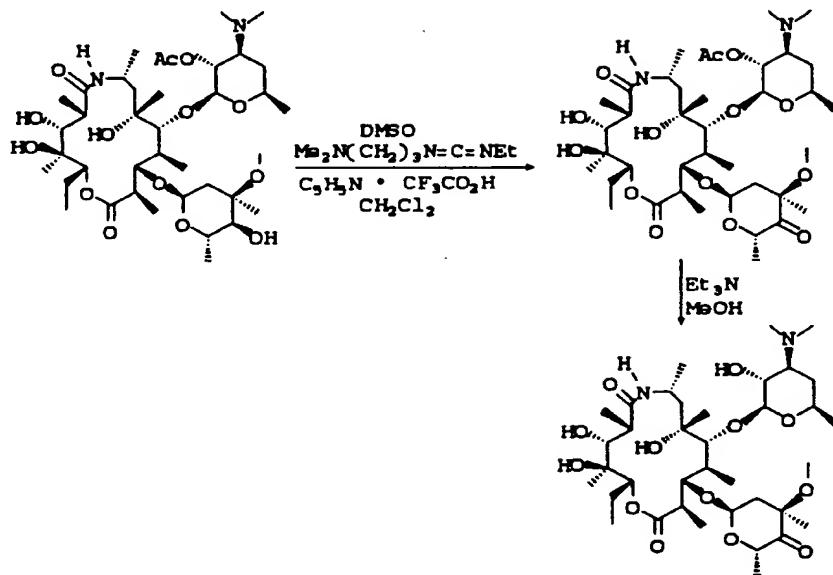
5

10

15

20

25



A solution of 2'-O-acetyl-8a-aza-8a-homoerythromycin A (1.45 g, 1.83 mmol) in dichloromethane (20 mL) was treated sequentially with dimethyl sulfoxide (1.3 mL, 18.4 mmol), 1-(3-dimethylaminopropyl)-3-ethylcarbodiimide (1.06 g, 5.52 mmol) and pyridinium trifluoroacetate (1.06 g, 5.52 mmol). The resulting solution was stirred at room temperature for 3.5 hours, then diluted with water (12 mL) and stirred while the pH of the aqueous phase was brought to 9.5 with aqueous sodium hydroxide. The layers were separated and the aqueous portion extracted with more dichloromethane (3 x 35 mL). The combined dichloromethane extracts were washed with 5% aqueous sodium bicarbonate, dried over magnesium sulfate, filtered and evaporated under vacuum to provide crude 2'-O-acetyl-4"-deoxy-4"-oxo-8a-aza-8a-homoerythromycin A as a foam.

<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.52 (d, NH), 5.35 (t, H-1''), 4.94 (dd, H-13), 4.72 (dd, H-2'), 4.37 (m, H-3 and H-5''), 4.31 (d, H-1'), 4.17 (m, H-8), 3.47 (d, H-5), 3.37 (m, H-5'), 3.43 (s, H-11), 3.32 (s, OCH<sub>3</sub>), 2.58 (m, H-2), 2.27 (br s, N(CH<sub>3</sub>)<sub>2</sub>), 2.06 (s, OCOCH<sub>3</sub>), 0.89 (t, CH<sub>2</sub>CH<sub>3</sub>).

Deacetylation was accomplished by dissolving the above product in methanol (12 mL), adding triethylamine (ca. 0.2 mL), and stirring the resulting solution at room temperature for 3 days. The solvent was removed under vacuum and the residue was chromatographed on EM silica gel 60 (230-400 mesh, 1.5 x 4.5 cm column packed under and eluted with 90:10:1 dichloromethane-methanol-conc. ammonium hydroxide, collecting six 25 mL fractions followed by 8 mL fractions). Fractions 11-15 were pooled and evaporated under vacuum to provide 4"-deoxy-4"-oxo-8a-aza-8a-homoerythromycin A (0.95 g) as a foam.

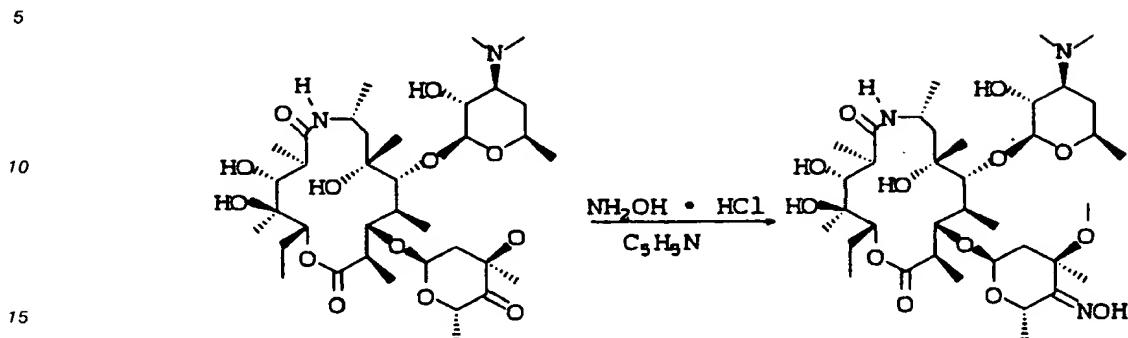
<sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 6.06 (br d, NH), 5.37 (t, H-1''), 4.92 (dd, H-13), 4.43 (m, H-3), 4.41 (q, H-5''), 4.21 (d, H-1'), 4.16 (m, H-8), 3.50 (d, H-5), 3.44 (s, H-11), 3.36 (m, H-5'), 3.29 (s, OCH<sub>3</sub>), 3.12 (dd, H-2'), 2.60 (dq, H-2), 2.42 (m, H-3'), 2.23 (s, N(CH<sub>3</sub>)<sub>2</sub>), 1.87 (m, H-4 and H-14a), 1.63 (m, H-4'eq), 0.87 (t, CH<sub>2</sub>CH<sub>3</sub>)

<sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 212.1, 177.8, 176.6, 103.7, 95.4, 83.2, 77.4, 77.3, 77.0, 76.8, 76.7, 74.7, 74.1, 71.6, 70.4, 70.0, 69.3, 65.4, 51.2, 45.5, 42.7, 42.6, 41.6, 40.5, 40.2, 36.8, 28.4, 27.6, 23.4, 21.6, 21.4, 20.8, 16.2, 16.1, 14.8, 11.3, 9.2.

FAB-MS m/z 748 (M + H), 591, 158.

## EXAMPLE 36

## 4"-Deoxy-4"-hydroxyimino-8a-aza-8a-homoerythromycin A



20 A mixture of 4"-deoxy-4"-oxo-8a-aza-8a-homoerythromycin A (0.90 g, 1.2 mmol), hydroxylamine hydrochloride (0.85 g, 12 mmol) and pyridine (4 mL) was stirred at room temperature for 24 hours, then evaporated under vacuum. The residue was stirred with water (25 mL) and dichloromethane (25 mL) while adjusting the pH of the aqueous phase from 5 to 11 with aqueous sodium hydroxide. The phases were separated and the aqueous portion was extracted with more dichloromethane (6 mL). The combined dichloromethane extracts were dried with magnesium sulfate, filtered, evaporated under vacuum, and three times dissolved in warm toluene (20 mL) and evaporated under vacuum to afford crude 4"-deoxy-4"-hydroxyimino-8a-aza-8a-homoerythromycin A (0.90 g) as a 3:1 mixture of oxime isomers.

25

30

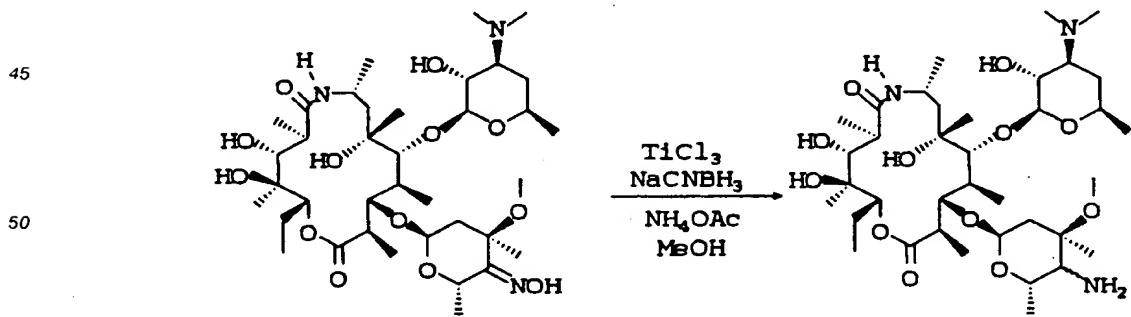
35 <sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 5.97 (br d, NH), 5.22 (t, H-1" of minor isomer), 5.14 (t, H-1" of major isomer), 5.07 (q, H-5" of major isomer), 4.95 (br d, H-13 of major isomer), 4.92 (br d, H-13 of minor isomer), 4.54 (q, H-5" of minor isomer), 4.38 (d, H-1'), 4.24 (m, H-3), 4.15 (m, H-8), 3.74 (d, H-5), 3.56 (m, H-5'), 3.45 (br s, H-11), 3.27 (s, OCH<sub>3</sub>), 3.16 (dd, H-2'), 3.47 (m, H-3'), 2.34 (s, N(CH<sub>3</sub>)<sub>2</sub>), 0.87 (t, CH<sub>2</sub>CH<sub>3</sub>).

40 <sup>13</sup>C NMR (CDCl<sub>3</sub>) δ 177.6, 176.3, 160.4, 157.4, 104.1, 102.9, 95.6, 95.4, 83.8, 83.2, 79.9, 76.9, 76.6, 75.0, 74.9, 74.2, 74.0, 70.6, 70.3, 69.4, 68.9, 66.8, 65.8, 65.5, 65.4, 51.1, 50.3, 45.3, 42.8, 42.4, 41.7, 41.6, 41.4, 40.7, 39.4, 35.3, 29.0, 28.9, 27.4, 27.3, 25.2, 23.4, 23.0, 21.7, 21.5, 21.4, 21.3, 17.5, 17.3, 16.5, 15.4, 14.8, 11.2, 11.1, 9.4.

FAB-MS m/z 785 (M + Na), 769 (M + Li), 763 (M + H), 592, 417, 158.

## EXAMPLE 37

## 4"-Deoxy-4"-amino-8a-aza-8a-homoerythromycin A



A solution of 4"-deoxy-4"-hydroxyimino-8a-aza-8a-homoerythromycin A (200 mg, 0.26 mmol) in methanol (4 mL) was treated sequentially with ammonium acetate (450 mg, 5.78 mmol), sodium

cyanoborohydride (48 mg, 95 % pure, 0.73 mmol) and titanium(III)chloride (0.50 mL of a 15% solution in 20% hydrochloric acid, 0.58 mmol). The resulting mixture was stirred at room temperature for 24.5 hours, then treated with nitromethane (8 drops) and stirred a few minutes. Evaporation of the solvent under vacuum left a residue which was stirred with dichloromethane and water while adjusting the pH of the aqueous phase to 9.5 with sodium hydroxide. The layers were separated and the aqueous portion was extracted with more dichloromethane. The combined organic extracts were dried with magnesium sulfate, filtered and evaporated under vacuum to a foam (163 mg).

5 The crude product was purified by chromatography on EM silica gel 60 (230-400 mesh, 1.5 x 30.5 cm column packed under and eluted with 90:10:1 dichloromethane-methanol-conc. ammonium hydroxide, 10 collecting 7 x 8 mL fractions then 43 x 3 mL fractions followed by 8 mL fractions). Fractions 53-62 were combined, evaporated under vacuum, and the residue was lyophilized from benzene (8 mL) containing a few drops of dichloromethane to afford 4"-deoxy-4"-amino-8a-aza-8a-homorythromycin A (76 mg) as a mixture of 4"-isomers (ca. 55:45 (4"R):(4"S) as determined by <sup>1</sup>H NMR).

15 <sup>1</sup>H NMR (CDCl<sub>3</sub>) δ 6.69 and 6.54 (two br d<sup>1</sup>s, NH), 5.08 and 5.06 (two overlapping d<sup>1</sup>s, H-1''), 4.87 (dd, H-13), 4.56 (q, H-5'' of 4"R-isomer), 4.50 (d, H-1' of 4"R-isomer), 4.44 (d, H-1' of 4"S-isomer), 4.30 (m, H-3), 4.13 (m, H-8), 3.99 (dq, H-5'' of 4"S-isomer), 3.55 and 3.53 (two overlapping d<sup>1</sup>s, H-5), 3.46 and 3.45 (two s<sup>1</sup>s, H-11), 3.26 (s, OCH<sub>3</sub> of 4"R-isomer), 3.24 (s, OCH<sub>3</sub> of 4"S-isomer), 3.16 and 3.13 (two overlapping dd<sup>1</sup>s, H-2'), 2.31 (br s, N(CH<sub>3</sub>)<sub>2</sub> of 4"S-isomer), 2.77 (br s, N(CH<sub>3</sub>)<sub>2</sub> of 4"R-isomer), 1.67 (m, H-4'eg), 1.48 (s, 6-CH<sub>3</sub>), 0.86 (t, CH<sub>2</sub>CH<sub>3</sub>).

20 <sup>13</sup>C NMR (CDCl<sub>3</sub>, 60 °C) δ 177.6, 177.5, 176.5, 176.3, 103.0, 102.6, 95.1, 94.9, 83.4, 83.0, 77.3, 77.2, 77.0, 76.8, 76.6, 75.1, 74.8, 74.1, 74.0, 72.7, 70.5, 70.2, 68.6, 68.4, 66.1, 65.8, 65.7, 62.8, 61.9, 57.9, 49.2, 45.7, 43.2, 43.0, 42.6, 42.5, 41.6, 41.5, 40.6, 40.5, 40.1, 35.1, 29.1, 29.0, 27.7, 23.2, 22.0, 21.7, 21.2, 18.7, 17.8, 16.2, 16.1, 14.6, 14.5, 11.0, 9.7, 9.6, 9.2, 9.1.

FAS-MS m/z 771 (M = Na), 592, 158.

25 The test procedures employed to measure the activity of the compounds of the invention are described below.

### EXAMPLE 38

30 The antibacterial activities of the compounds of Examples 8 and 10 against a panel of aerobic Gram positive and negative bacteria is shown in the following Table. The assay employs a liquid turbidimetric microtiter method for the determination of the minimum inhibitory concentration (MIC) in broth media. The MIC endpoint in mcg/ml is defined as the lowest concentration of test compound that completely inhibits the growth (absence of turbidity) of bacteria. The MIC is generally not an absolute value but rather a 35 concentration range that falls within a two-fold dilution limit. Generally twelve two-fold dilutions of the test compound are employed with the initial concentration set at 128 mcg/ml.

40

45

50

55

TABLE I5 In Vitro Activity of Compounds of Examples 9 and 10.

10 MIC Values (mcg/ml)

15 Microorganism (9) (10)

Enterococcus faecalis	MB 5407	8	4
Enterococcus faecium	MB 5416	0.25	0.25
Streptococcus agalactiae	CL 1343	≤0.06	≤0.06
Staphylococcus aureus	MB 2865	0.5	0.5
Staphylococcus epidermidis	MB 5414	1	1
Staphylococcus haemolyticus	MB 5412	0.5	0.5
Streptococcus pneumoniae	CL 2883	≤0.06	≤0.06
Streptococcus pyogenes	MB 2874	≤0.06	≤0.06
Streptococcus pyogenes	MB 54061	128	128
Streptococcus viridans	CL 2943	8	1
Escherichia coli	MB 2884	4	1
Escherichia coli	MB 4926	2	0.06
Klebsiella pneumoniae	MB 4005	8	2
Pseudomonas stutzeri	MB 1231	≤0.06	≤0.06

35 Example (9) 9-Deoxo-8a-aza-8a-homoerythromycin A  
Example (10) 9-Deoxo-8a-methyl-8a-aza-8a-homoery-  
thromycin A

40

45

50

55

TABLE II

In vitro Activity of Compounds of Examples (15), (17), (19), and (20)						
	Microorganism	15	17	19	20	Erythromycin A
5	Enterococcus faecalis	MB 5407	1	4	2	2
10	Staphylococcus aureus	MB 2865	0.5	1	1	0.12
15	Staphylococcus epidermidis	MB 5414	0.25	0.5	0.51	0.12
20	Streptococcus pneumoniae	CL 2883	≤0.06	≤0.06	≤0.06	≤0.06
25	Streptococcus pyogenes	MB 2874	≤0.06	≤0.06	≤0.06	≤0.06
30	Enterobacter cloacae	CL 4298	0.5	≤0.06	≤0.06	16
35	Escherichia coli	MB 2884	1	0.12	0.12	16
40	Klebsiella pneumoniae	MB 4005	1	0.12	0.12	32
45	Haemophilus influenzae	MB 5363	0.5	0.5	0.25	2
50	Haemophilus influenzae	AT 43163	0.5	0.25	0.25	2

Example 15 4"-Epi-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

Example 17 4"-Deoxy-4"-amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A (2:1 mixture of 4"-isomers)

Example 19 4"-Deoxy-4"(S)-amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

Example 20 4"-Deoxy-4"(R)-amino-9-deoxo-8a-aza-8a-methyl-8a-homoerythromycin A

The compounds of formula (II) are useful as an antibacterial agents both in vitro and in vivo, and their spectrum of activity is similar to that of erythromycin A. Consequently, they can be used for the same purposes, and, in the same manner as erythromycin A. In general, the antibacterial compounds of formula II and salts thereof, exhibit in vitro activity against a variety of Gram-positive microorganisms, e.g. Streptococcus pyogenes, and against certain Gram-negative microorganisms such as those of spherical or ellipsoidal shape (cocci). Their activity is readily demonstrated by in vitro tests against various micro-organisms. Their in vitro activity renders them useful for topical application; for sterilization purposes, e.g., sick-room utensils; and as industrial antimicrobials, for example, in water treatment, slime control, and preservation of paint and wood. The extrapolation of such in vitro tests to support for such utilities for macrolide compounds is taught in U.S. Patent No. 4,518,590, cited above.

While the method of the invention has been described, exemplified and illustrated in reference to certain preferred embodiments thereof, those skilled in the art will appreciate that various changes, modifications and substitutions can be made therein without departing from the spirit and scope of the invention.

It is intended, therefore, that the invention be limited only by the scope of the claims which follow and that such claims be interpreted as broadly as is reasonable.

#### 40 Claims

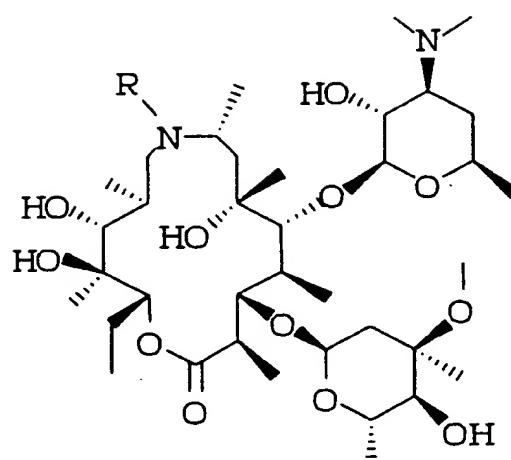
1. A method of making 4" aminated derivatives of 9-deoxo-8a-aza-8a-alkyl-8a-homoerythromycin A, comprising the steps of
  - (a) oxidizing a compound of the formula

45

50

55

5



10

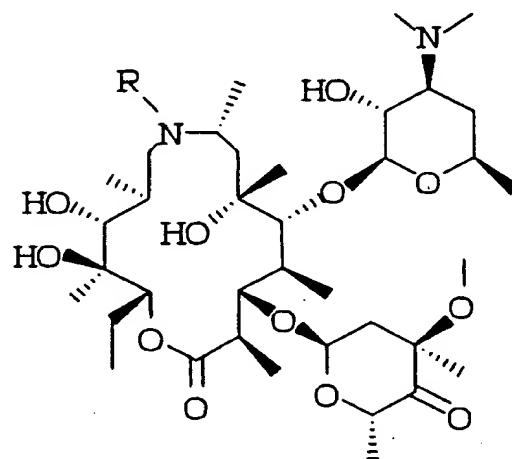
15

wherein R is hydrogen, hydroxyl, C<sub>1</sub>-C<sub>10</sub>alkyl, or C<sub>1</sub>-C<sub>10</sub>alkoxy-carbonyl C<sub>1</sub>-C<sub>10</sub>alkyl to produce a compound of the formula

25

30

35



40

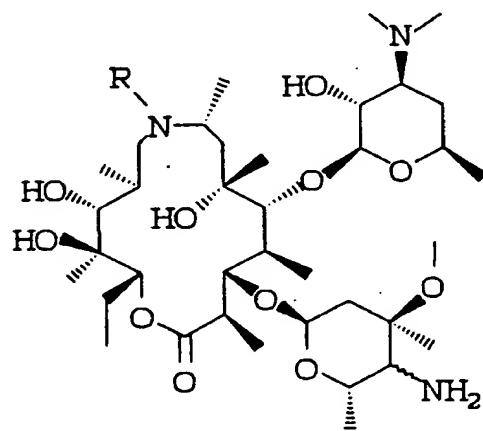
and

(b) aminating the product of step (a) to produce the product compound of the formula

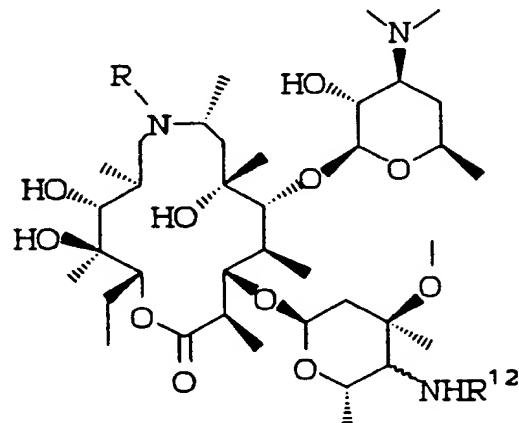
45

50

55



2. The process as claimed in Claim 1, comprising the additional step of substituting the product of step (b) therein to produce the product compound of the formula



wherein  $R^{12}$  is

$C_{1-10}$ alkylcarbonyl,  
 $C_{1-10}$ alkoxycarbonyl,  
arylcarbonyl,  
aryl  $C_{1-10}$ alkylcarbonyl or  
aryl  $C_{1-10}$ alkoxycarbonyl.



European Patent  
Office

## EUROPEAN SEARCH REPORT

**Application Number**

EP 92 20 3905

DOCUMENTS CONSIDERED TO BE RELEVANT			
Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int. Cl.5)
A	WO-A-9 011 288 (ABBOTT LABORATORIES) * abstract * * page 18, line 4 - page 41, line 4 * ---	1, 2	C07H17/08 A61K31/70
A	EP-A-0 345 627 (ABBOTT LABORATORIES) * abstract * * page 6, line 1 - page 7, line 50 * ---	1, 2	
A	EP-A-0 109 253 (PFIZER INC.) * the whole document * ---	1, 2	
P, X	EP-A-0 508 699 (MERCK AND CO. INC.) * the whole document * -----	1, 2	
TECHNICAL FIELDS SEARCHED (Int. Cl.5)			
C07H A61K			
The present search report has been drawn up for all claims			
Place of search	Date of completion of the search	Examiner	
THE HAGUE	02 MARCH 1993	SCOTT J.R.	
CATEGORY OF CITED DOCUMENTS			
X : particularly relevant if taken alone	T : theory or principle underlying the invention		
Y : particularly relevant if combined with another document of the same category	E : earlier patent document, but published on, or after the filing date		
A : technological background	D : document cited in the application		
O : non-written disclosure	L : document cited for other reasons		
P : intermediate document	& : member of the same patent family, corresponding document		

**This Page Blank (uspto)**